Investigating brain activity prior to tics and modulating this activity with peripheral nerve stimulation to suppress tics in Tourette syndrome

Bàrbara Morera Màiquez, BSc., MSc.

Thesis abstract

Tourette syndrome (TS) is a neurological disorder characterized by motor and vocal tics. It is associated with a dysfunction in the cortical-striatal-thalamic-cortical (CSTC) circuit that leads to an overexcited motor cortex resulting in the execution of the motor or vocal action (tics). Most individuals report having uncomfortable sensations or premonitory urges (PU) preceding some of their tics, and many individuals report that their tics result from these sensations. Tic suppression is associated with an inhibited motor cortex and it is present during voluntary movements in order to perform the desired motor action.

The first aim of this thesis was to explore the brain activity preceding tics and brain activity preceding voluntary movement. To investigate this, I used electroencephalography (EEG) and functional magnetic resonance imaging (fMRI) to explore the brain activity preceding tics, and I used EEG to examine brain activity preceding voluntary movements in both healthy subjects and subjects with TS. Time-frequency analyses showed no consistent pattern of cortical activity preceding tics, although individual tics were characterized by the presence of an increased excitability in the sensorimotor cortex (SMC) before their execution; and BOLD activity showed activation in motor and sensory brain areas preceding tics, previously reported in other studies. Time-frequency analyses showed a reduced excitability in the SMC before voluntary movements in subjects with TS.

The second aim was to investigate the use of a non-invasive brain stimulation technique to modulate this cortical excitability. I used median nerve stimulation (MNS) while recording EEG to explore whether rhythmic MNS entrains brain oscillations in the SMC, then I used rhythmic MNS during a motor task to explore its effects on voluntary movement, finally I used rhythmic MNS on TS to explore its effects on tics. Time-frequency analyses and inter-trial coherence showed clear evidence that rhythmic MNS entrains brain oscillations in the sensorimotor cortex; response times in a computer motor task showed a reduction of speed in motor response during rhythmic MNS; video recordings showed a decrease in the number of tics during MNS.

Acknowledgements

First of all, I would like to thank my supervisors Stephen Jackson and Georgina Jackson for their incredibly great support and guidance throughout my PhD. I'm extremely grateful for all the trust you have put on me since the beginning and all the knowledge and skills I've acquired because of that.

I would also like to thank other members from the Prof. Jackson's lab and friends: Katherine Dyke, thank you for your help with Matlab during my first steps of coding; Hilmar Siggurdson, thank you for your help with settling in at the beginning of my PhD, problem-solve Matlab based problems, and for training me to be an MRI assistant; Jennifer Salvage and Mairi Houlgreave, thanks for your peer support. Even though my TMS studies didn't make it into my thesis, I would also like to thank Soyoung Kim, ex-member of the team, for helping me and teaching me how to use TMS.

Also, Miguel Santos, thank you for training me on how to set up and collect EEG data; Paul Morgan, thanks for setting up the scanner protocol and helping with finding the optimal attachment of the MRI compatible camera onto the head coil; Joseph Kilgariff thanks for training me to administer the YGTSS questionnaire.

Very importantly, I would like to thank all the children, adolescents and adults who participated in my studies. It wouldn't have been possible to achieve this without you.

Lastly, Joe Vallender, thank you for always paying attention when practicing my presentations with you, and for your constant support and encouragement and my family, thank you, I wouldn't have made it without you.

Table of contents

THESIS ABSTRACT ..... **II**

ACKNOWLEDGEMENTS ..... **III**

TABLE OF CONTENTS ..... **IV**

TABLE OF FIGURES ..... **VI**

TABLE OF TABLES ..... **XI**

CHAPTER 1 - THESIS OVERVIEW AND GENERAL INTRODUCTION TO TOURETTE SYNDROME AND ENTRAINMENT OF NEURAL OSCILLATIONS **1**

1.1. THESIS OVERVIEW 1

1.2. GILLES DE LA TOURETTE SYNDROME 3

1.3. ENTRAINMENT OF BRAIN OSCILLATIONS 11

1.4. SUMMARY AND RESEARCH AIM 14

CHAPTER 2 - GENERAL METHODS **15**

2.1. PARTICIPANTS AND CLINICAL MEASUREMENTS 15

2.2. ELECTROENCEPHALOGRAPHY (EEG) 18

2.3. MAGNETIC RESONANCE IMAGING (MRI) 26

2.4. MEDIAN NERVE STIMULATION (MNS) 33

CHAPTER 3 - EXPLORING THE BRAIN ACTIVITY PRIOR TO TICS AND VOLUNTARY MOVEMENTS USING EEG **38**

CHAPTER 3A.1 - EXPLORING THE BRAIN ACTIVITY PRIOR TO VOLUNTARY MOVEMENTS **38**

3A.1.1. INTRODUCTION 38

3A.1.2. METHODS 40

3A.1.3. RESULTS 45

3A.1.4. DISCUSSION 52

3A.1.5. CONCLUSION AND CONTINUANCE 54

CHAPTER 3A.2 - BRAIN ACTIVITY PRIOR TO TICS **55**

3A.2.1. INTRODUCTION 55

3A.2.2. METHODS 56

3A.2.3. RESULTS 60

3A.2.4. DISCUSSION 65

3A.2.5. CONCLUSION 68

CHAPTER 3B - EXPLORING THE BRAIN ACTIVITY PRIOR TO A TIC USING FMRI **69**

3B.1. INTRODUCTION 69

3B.2. METHODS 71
3B.3. RESULTS 78
3B.4. DISCUSSION 84
3B.5. CONCLUSION 86

CHAPTER 4 - EXPLORING MNS AS A TOOL TO ENTRAIN THE MOTOR CORTEX 88

4.1. INTRODUCTION 88
4.2 MU ENTRAINMENT STUDY 90
4.3. BETA ENTRAINMENT STUDY 104
4.4. DISCUSSION 118
4.5. CONCLUSION AND CONTINUANCE 122

CHAPTER 5 - EXPLORING MNS BEHAVIOURAL EFFECTS OF MU ENTRAINMENT 124

5.1. INTRODUCTION 124
5.2. METHODS 125
5.3. RESULTS 128
5.4. DISCUSSION 132
5.5. CONCLUSION AND CONTINUANCE 135

CHAPTER 6 - EXPLORING MNS CLINICAL EFFECTS OF MU ENTRAINMENT 136

6.1. INTRODUCTION 136
6.2. METHODS 137
6.3. RESULTS 142
6.4. DISCUSSION 147
6.5. CONCLUSION 150

CHAPTER 7 - GENERAL DISCUSSION AND THESIS CONCLUSION 151

7.1. ACTIVITY IN THE BRAIN BEFORE TICS 151
7.2. MODULATION OF THE BRAIN ACTIVITY IN THE MOTOR CORTEX AS A TREATMENT FOR TS 153
7.3. THESIS LIMITATIONS AND FUTURE WORK 155
7.4. CONCLUSION 157

REFERENCES 158

APPENDIX 1 167

APPENDIX 2 174

Table of Figures

Figure 2.1. Channel locations with the name of the electrodes 22

Figure 2.2 Shape of the HRF. ID: initial dip; TP: Time of the stimulus until peak; H: height of response; W: width of the HRF at half the height; PSU: post-stimulus undershoot. This image has been taken from The Handbook of functional MRI Data analysis..... 28

Figure 2.3. Scheme showing the process of MRI image registration 29

Figure 2.4. Illustration of cluster-level inference. Those clusters over the cluster-forming threshold (uc) are found significant. This image has been taken from The Handbook of functional MRI Data analysis. 30

Figure 2.5. Image showing the course of the median nerve from the finger tips to the brachial plexus. This figure has been taken from the website teachmeanatomy 34

Figure 2.6. Image showing the placement of the electrodes..... 37

Figure 3a.1.1. ERP shown in the contralateral M1. Time 0 indicates the button press. 46

Figure 3a.1.2. ERP shown in the contralateral M1. Time 0 indicates the button press. 46

Figure 3a.1.3. A: Time-frequency analyses from the contralateral M1 in the mu band; B: Values of power at 12Hz recorded from the contralateral M1 in time. Time 0 indicates the button press. The black horizontal line indicates the time points were the values of power are significant. 47

Figure 3a.1.4. A: Time-frequency analyses from the contralateral M1 in the mu band; B: Values of power at 12Hz recorded from the contralateral M1 in time. Time 0 indicates the start of the movement. 48

Figure 3a.1.5. A: Time-frequency analyses from the contralateral M1 in the beta band; B: Values of power at 19Hz recorded from the contralateral M1 in time. Time 0 indicates the start of the movement. The black horizontal line indicates the time points were the values of power are significant. 49

Figure 3a.1.6. A: Time-frequency analyses from the contralateral M1 in the beta band; B: Values of power at 19Hz recorded from the contralateral M1 in time. Time 0 indicates the start of the movement. The black horizontal line indicates the time points were the values of power are significant. 50

Figure 3a.1.7. A: Relationship between the Mu power prior to button press and the global YGTSS scores. B: Relationship between the Beta power prior to button press and the global YGTSS scores. 51

Figure 3a.1.8. A: Relationship between the Mu power at the time of button press and the global YGTSS scores. B: Relationship between the Beta power at the time of button press and the global YGTSS scores. 52

Figure 3a.2.1. Average ERP from the average electrodes on the SMC. Red line indicates the start of the tic. 61

Figure 3a.2.2. Average ERP from the average electrodes on the SMC for each subject. Red line indicates the onset of the tic. Plots also show the number of trials (in grey) for each participant. PU: PUTS-R score..... 62

Figure 3a.2.3. Time-frequency plots in alpha band (8-13Hz) for the average electrodes on the SMC. 0 indicates the onset of the tic..... 63

Figure 3a.2.4. ERSP values for the SMC in alpha band. 0 indicates the onset of the tic. 63

Figure 3a.2.5. Time-frequency plots in beta band (13-30Hz) for the average electrodes on the SMC. 0 indicates the onset of the tic..... 64

Figure 3a.2.6. ERSP values for the SMC in alpha band. 0 indicates the onset of the tic. 64
Figure 3a.2.7. Average of trials rank-ordered to the strongest ERD peak before tic onset in alpha band (left) and beta band (right) for the average electrodes on the SMC. 0 indicates the onset of the tic 65
Figure 3b.1. Clusters exhibiting a significant BOLD increase corresponding to two seconds before tic onset (p<0.05 corrected for FDR) in whole-brain analyses 81
Figure 3b.2. Clusters exhibiting a significant BOLD increase corresponding to one second before tic onset (p<0.05 corrected for FDR) in whole-brain analyses 82
Figure 3b.3. Clusters exhibiting a significant BOLD increase corresponding to tic onset (p<0.05 corrected for FDR) in whole-brain analyses 83
Figure 4.1. Scheme of the design of the study 91
Figure 4.2. Time frequency analyses recorded from electrode C1 for A: the arrhythmic condition, B: the rhythmic condition, and C: the subtraction of the arrhythmic condition from the rhythmic condition. 0 indicates the time of the first pulse of the train. The black rectangle indicates W1 at 12Hz frequency and the red rectangle indicates W2 at 12Hz frequency. 93
Figure 4.3. Time-frequency values recorded from electrode C1 at 12Hz for both the rhythmic (red) and arrhythmic (blue) conditions. 0 indicates the time of the first pulse of the train. Black lines indicate the time points where the time-frequency values between both conditions were significantly different. The black rectangle indicates W1 and the red rectangle indicates W2. 94
Figure 4.4. ITC analyses recorded from electrode C1 for A: the arrhythmic condition, B: the rhythmic condition, and C: the subtraction of the arrhythmic condition from the rhythmic condition. 0 indicates the time of the first pulse of the train. The black rectangle indicates W1 at 12Hz frequency and the red rectangle indicates W2 at 12Hz frequency. 95
Figure 4.5. ITC values recorded from electrode C1 at 12Hz for both the rhythmic (red) and arrhythmic (blue) conditions. 0 indicates the time of the first pulse of the train. Black line indicates the time points where the ITC values between both conditions are significantly different. The black rectangle indicates W1 and the red rectangle indicates W2. 96
Figure 4.6. Time frequency analyses of the rhythmic condition for A: electrode C1, B: electrode C2, and C: the subtraction of the electrode C2 from the electrode C1. 0 indicates the time of the first pulse of the train. The black rectangle indicates W1 at 12Hz frequency and the red rectangle indicates W2 at 12Hz frequency 97
Figure 4.7. Time-frequency values recorded from electrode C1 (red) and electrode C2 (blue) at 12Hz for the rhythmic condition. 0 indicates the time of the first pulse of the train. Black lines indicate the time points where the time-frequency values between both electrodes are significantly different. The black rectangle indicates W1 and the red rectangle indicates W2. 98
Figure 4.8. ITC analyses of the rhythmic condition recorded from A: electrode C1, B: electrode C2, and C: the subtraction of the electrode C2 from the electrode C1. 0 indicates the time of the first pulse of the train. The black rectangle indicates W1 and the red rectangle indicates W2. 99
Figure 4.9. ITC values recorded from electrode C1 (red) and electrode C2 (blue) at 12Hz in the rhythmic condition. 0 indicates the time of the first pulse of the train. Black line indicates the time points where the ITC values

between electrodes are significantly different. The black rectangle indicates W1 and the red rectangle indicates W2. 100
Figure 4.10. Beta oscillations (8-13Hz) for both rhythmic (red) and arrhythmic (blue) conditions for each pulse of the train. 101
Figure 4.11 Scalp topographies averaged for A: pulses 1-3 and B: pulses 8-10 in the arrhythmic (left) and rhythmic (center) conditions. Scalp maps on the right show the electrodes that showed a significant difference in voltage between conditions. 102
Figure 4.13. Time-frequency values for electrode C1 at the average frequency of 13-30Hz for both the rhythmic (red) and arrhythmic (blue) conditions. 0 indicates the time of the first pulse of the train. Black lines indicate the time points where the time-frequency values between both conditions are significantly different. The black rectangle indicates W1 and the red rectangle indicates W2. 103
Figure 4.12. Time frequency analyses for electrode C1 for the rhythmic condition (left) and the arrhythmic condition (right). 0 indicates the time of the first pulse of the train. The vertical dotted line indicates the end of the train. The white rectangle indicates beta band (13-30Hz) after the stimulation. 103
Figure 4.14. Scheme of the design of the study 105
Figure 4.15. Time frequency analyses for electrode C1 for A: the arrhythmic condition, B: the rhythmic condition, and C: the subtraction of the arrhythmic condition from the rhythmic condition. 0 indicates the time of the first pulse of the train. The rectangle indicates the 18-20Hz frequency for the whole length of the train .. 107
Figure 4.16. Time-frequency values for electrode C1 at the average frequency of 18-20Hz for both the rhythmic (red) and arrhythmic (blue) conditions. 0 indicates the time of the first pulse of the train. Black line indicates the time points where the time-frequency values between both conditions are significantly different. The rectangle indicates the length of the train 108
Figure 4.17. ITC analyses for electrode C1 for A: the arrhythmic condition, B: the rhythmic condition, and C: the subtraction of the arrhythmic condition from the rhythmic condition. 0 indicates the time of the first pulse of the train. The rectangle indicates the 18-20Hz frequency for the whole length of the train 109
Figure 4.18. ITC values for electrode C1 at the average frequency of 18-20Hz for both the rhythmic (red) and arrhythmic (blue) conditions. 0 indicates the time of the first pulse of the train. Black line indicates the time points where the ITC values between both conditions are significantly different. The rectangle indicates the whole length of the train 110
Figure 4.19. Time frequency analyses of the rhythmic condition for A: electrode C1, B: electrode C2, and C: the subtraction of the electrode C2 from the electrode C1. 0 indicates the time of the first pulse of the train. The rectangle indicates the 18-20Hz frequency for the whole length of the train 111
Figure 4.20. Time-frequency values for electrode C1 (red) and electrode C2 (blue) at the average frequency of 18-20Hz for the rhythmic conditions. 0 indicates the time of the first pulse of the train. Black line indicates the time points where the time-frequency values between both electrodes are significantly different. The rectangle indicates the whole length of the train 112

Figure 4.21. Inter-trial coherence analyses of the rhythmic condition for A: electrode C1, B: electrode C2, and C: the subtraction of the electrode C2 from the electrode C1. 0 indicates the time of the first pulse of the train. The rectangle indicates the 18-20Hz frequency for the whole length of the train 113
Figure 4.22. ITC values for electrode C1 (red) and electrode C2 (blue) at the average frequency of 18-20Hz for the rhythmic condition. 0 indicates the time of the first pulse of the train. Black line indicates the time points where the ITC values between electrodes are significantly different. The rectangle indicates the whole length of the train 114
Figure 4.23. Beta oscillations (13-30Hz) for both rhythmic (red) and arrhythmic (blue) conditions for each pulse of the train. 115
Figure 4.24. Scalp topographies averaged for A: pulses 1-3 and B: pulses 8-10 in the arrhythmic (left) and rhythmic (center) conditions. Scalp maps on the right show the electrodes that showed a significant difference in voltage between conditions. 116
Figure 4.25. Time frequency analyses for electrode C1 for the rhythmic condition (left) and the arrhythmic condition (right). 0 indicates the time of the first pulse of the train. The vertical dotted line indicates the end of the train 117
Figure 4.26. Time-frequency values for electrode C1 at the average frequency of mu (8-13Hz) for both the rhythmic (red) and arrhythmic (blue) conditions. 0 indicates the time of the first pulse of the train. Black line indicates the time points where the time-frequency values between both conditions are significantly different. The rectangle indicates the whole length of the train 117
Figure 4.27. Time-frequency values for electrode C1 at the average frequency of 21-30Hz for both the rhythmic (red) and arrhythmic (blue) conditions. 0 indicates the time of the first pulse of the train. Black line indicates the time points where the time-frequency values between both conditions are significantly different. The rectangle indicates the whole length of the train 118
Figure 5.1. Scheme of the design of the study 126
Figure 5.2. Boxplot of the RT for each condition. Every dot represents one subject's average RT 129
Figure 5.3. Mean RT for each bin and the corresponding error percentage for the rhythmic (red) and arrhythmic trials (blue) 130
Figure 5.4. Boxplot of the RT for each condition. Every dot represents one subject's average RT 131
Figure 5.5. Mean RT for each bin and the corresponding error percentage for the rhythmic (red) and arrhythmic trials (blue) 132
Figure 6.1. Scheme of the design of the study 140
Figure 6.2. Average number of tics in each condition. The plot also shows the number of tics in each participant at each condition 143
Figure 6.3. Average intensity of tics in each condition. The plot also shows the number of tics in each participant at each condition 144
Figure 6.4. Average of the ratings in intensity of the urge to tic in each condition. The plot also shows the number of tics in each participant at each condition 145
Figure 8.1. ERP shown in the contralateral M1. Time 0 indicates the start of the button press. 168

FIGURE 8.2. ERP SHOWN IN THE CONTRALATERAL M1. TIME 0 INDICATES THE BUTTON PRESS. 169
FIGURE 8.3. A: TIME-FREQUENCY ANALYSES FROM THE CONTRALATERAL M1 IN THE MU BAND; B: VALUES OF POWER AT 12HZ RECORDED FROM THE CONTRALATERAL M1 IN TIME. TIME 0 INDICATES THE BUTTON PRESS. THE BLACK HORIZONTAL LINE INDICATES THE TIME POINTS WERE THE VALUES OF POWER ARE SIGNIFICANT. 170
FIGURE 8.4. A: TIME-FREQUENCY ANALYSES FROM THE CONTRALATERAL M1 IN THE MU BAND; B: VALUES OF POWER AT 12HZ RECORDED FROM THE CONTRALATERAL M1 IN TIME. TIME 0 INDICATES THE START OF THE MOVEMENT. 171
FIGURE 8.5. A: TIME-FREQUENCY ANALYSES FROM THE CONTRALATERAL M1 IN THE BETA BAND; B: VALUES OF POWER AT 19HZ RECORDED FROM THE CONTRALATERAL M1 IN TIME. TIME 0 INDICATES THE START OF THE MOVEMENT. THE BLACK HORIZONTAL LINE INDICATES THE TIME POINTS WERE THE VALUES OF POWER ARE SIGNIFICANT. 172
FIGURE 8.6. A: TIME-FREQUENCY ANALYSES FROM THE CONTRALATERAL M1 IN THE BETA BAND; B: VALUES OF POWER AT 19HZ RECORDED FROM THE CONTRALATERAL M1 IN TIME. TIME 0 INDICATES THE START OF THE MOVEMENT. THE BLACK HORIZONTAL LINE INDICATES THE TIME POINTS WERE THE VALUES OF POWER ARE SIGNIFICANT. 173
FIGURE 9.1. TIME-FREQUENCY PLOTS IN ALPHA, BETA AND GAMMA BANDS (8-63HZ) FOR THE AVERAGE ELECTRODES ON THE SMC. 0 INDICATES THE ONSET OF THE TIC. 174

Table of Tables

Table 3A.1.1. Participant characteristics 42

Table 3A.2.1. Participant characteristics 58

Table 3A.2.2 Participant ID with their corresponding number of trials 61

Table 3B.1. Participant characteristics 73

Table 3B.2. Collected data 79

Table 3B.3. Number of facial tics and number of button presses associated with urges in each run 80

Table 3B.4. MNI coordinates and corresponding brain regions of statistically significant clusters of activation associated with button presses (p<0.05 corrected for FDR). Z-Max indicates the maximum z-score within the brain region. 81

Table 3B.5. MNI coordinates and corresponding brain regions of statistically significant clusters of activation associated with button presses (p<0.05 corrected for FDR). Z-Max indicates the maximum z-score within the brain region 82

Table 3B.6. MNI coordinates and corresponding brain regions of statistically significant clusters of activation associated with button presses (p<0.05 corrected for FDR). Z-Max indicates the maximum z-score within the brain region. 83

Table 6.1. Participant characteristics 139

CHAPTER 1

Chapter 1 - Thesis overview and general introduction to Tourette Syndrome and entrainment of neural oscillations

---

1.1. Thesis overview

Within this thesis I used neuroimaging techniques to explore brain activity preceding voluntary movement and tics in people with Tourette syndrome (TS) and used non-invasive brain stimulation to modulate this brain activity and generate an effect on the execution of voluntary movements and the execution of tics. Briefly, I used EEG to examine the activity in the brain before voluntary movement in healthy subjects (HS) and subjects with TS (Chapter 3.a1); then I examined the activity in the brain before tics using EEG (Chapter 3.a2) and fMRI (Chapter 3.b). After that, I used median nerve stimulation (MNS) to modulate this brain activity in the motor cortex (Chapter 4) and showed the effect of this modulation on motor behaviour in HS (Chapter 5) and tic execution in subjects with TS (Chapter 6).

The main background research that leads to the research in this thesis is that cortical excitability plays an important role in movement, tic execution and premonitory urges. Chapter 3a1 examined this on a study exploring motor cortical excitability before voluntary movement in HS and subjects with TS. This was done by asking participants to perform a Go/NoGo task on a computer while EEG was being recorded. Results showed an increase in excitability of the motor cortex in both the mu and beta band in HS but only in the beta band in TS, prior and during movement execution. Chapter 3a2 explored motor cortical excitability before tics. This was done by recording EEG while participants rested and let their tics happen. Results did not show an increase in excitability immediately before tics when tics were analysed as a group but they did show an increase in excitability within the second before the tic

execution in both mu and beta bands. Chapter 3b explored the brain activity at the moment of strongest feeling of urge to tic as well as before tic execution, using fMRI. Participants were placed inside the scanner and pressed a button whenever they felt a strong urge to tic. Participant's facial tics were recorded with an MRI compatible camera and a researcher standing next to the participant pressed a button whenever she saw a tic on the body of the participant. Whole brain analyses were performed at tic onset, 1 second before tic onset, 2 seconds before tic onset, and at the participant's strong urge to tic. Results showed activity in the brain before tics, which included activity in the motor and somatosensory cortices, but not at the participant's strong urge to tic.

In Chapter 4, I delivered rhythmic and arrhythmic electric pulses using MNS on HS while recording EEG to examine the effects of the stimulation on cortical activity. This was done in two different studies; in one study stimulation was delivered at a frequency in the mu band and in the second study stimulation was delivered at a frequency in the beta band. Time-frequency analyses and inter-trial coherence analyses between the rhythmic and arrhythmic stimulations in the contralateral motor cortex showed that the rhythmic stimulation evoked an increase in power over the motor cortex at the stimulated frequency that did not happen when delivering arrhythmic stimulation. Increase in power is known to be indicative of decrease in cortical excitability. Plots of the brain oscillations at the stimulated frequency band suggested that the effect is generated by the phase resetting of the oscillations produced by the stimulation. Overall, the main finding of these two studies is that rhythmic MNS modulates the motor cortex by decreasing its excitability. In the next two chapters I examined the effect of the rhythmic MNS on motor behaviour and on tic execution.

Chapter 5 explored the effects of the rhythmic MNS on motor behaviour in HS. In the study, participants received either rhythmic or arrhythmic stimulation while having to initiate a motor response to a stimulus in a computer task. Results showed that rhythmic stimulation reduces the time of motor response compared to arrhythmic stimulation.

Finally, Chapter 6 explored the effects of the rhythmic MNS on the execution of tics in subjects with TS. In the study, participants received either constant rhythmic stimulation, constant arrhythmic stimulation or no stimulation at all while having to rate the intensity of their urges using an electronic slider. This was done while participants were being video recorded. To analyse the data, tics were counted, their intensity was rated, and the output of the slider was measured. Results showed an overall decrease on tic execution, an overall decrease on tic intensity and an overall decrease in urge intensity.

1.2. Gilles de la Tourette syndrome

Tourette syndrome is a neurological disorder characterized by the childhood onset of both motor and vocal tics, lasting for at least one year. Tics are involuntary, repetitive, stereotyped behaviours that occur with a limited duration. It is estimated to have an international prevalence between 0.3 and 0.9% in schoolchildren (Scharf et al., 2015), being more common in males than females. Motor and vocal tics vary from simple to complex. Simple motor tics involve a single contraction, such as head jerk, and complex motor tics involve contractions of groups of muscles, such as turning and twisting of neck muscles; simple vocal tics refers to a single sound, such as grunting or sniffing, and complex vocal tics can involve uttering words or phrases and can consist of repetitive sounds, words or utterances (palilalia), the production of inappropriate or obscene utterances (coprolalia), or the repetition of another's words (echolalia). Tics often appear at the age between 5-7 and peaks at about 10-12 years of age; then, in early adulthood, the majority of patients gain control over their symptoms, while in others symptoms continue to persist, and even increase in intensity and severity (Leckman, 2002).

TS can be presented alone ("pure" TS), but it is often presented with one or more comorbidities, which occurs up to 90% of the cases (Wright, Rickards, & Cavanna, 2012). The most common comorbidities are attention deficit hyperactivity disorder (ADHD), obsessive compulsive disorder (OCD) and

ADHD and OCD together, but other impulse control disorders are also common (Wright et al., 2012).

Tic severity can be measured with the YGTSS Global Tic Severity Scale (YGTSS). It is based on the rates of severity of motor and vocal tics in number, frequency, intensity, complexity, and interference, as well as the rating of distress and impairment experienced in the daily life in the interpersonal, academic, and occupational environment. More detail about the YGTSS is found on Chapter 2.

Some individuals with TS report having uncomfortable sensations preceding a tic. These sensations are defined as an urge, itch, impulse, pressure, tension or tingle (Libraries, 2006) in the joints, bones, muscles, and other parts of the body (Steinberg et al., 2010). After the person tics, there is usually a temporary relief of these sensations until they appear again. It is usually compared to sneezing (an uncomfortable sensation is felt with the need to sneeze, which is relieved after sneezing until there is the need to sneeze again). These sensations are called premonitory urges (PU), but they can also be referred as sensory phenomena. These sensations are reported by young adults but not usually by children under 10 years of age (Leckman, Walker, & Cohen, 1993). Some individuals take advantage of these sensations in order to temporally suppress their tics. Although the suppression of tics emerge with the appearance of PU, not all the individuals able to suppress tics report having PU, suggesting that these two processes may be independent (Banaschewski & Rothenberger, 2003). PU severity can be measured with the Premonitory Urge for Tics Scale (PUTS), which uses 9 items measuring sensory and mental phenomena. More detail about the PUTS is found on Chapter 2.

A review of the current knowledge of the syndrome's management stated that tics can cause subjective discomfort (such as pain or injury), sustained social problems (such as social isolation or bullying), social and emotional problems (such as reactive depressive symptoms), or functional interference (such as impairment of academic achievements) (Cavanna & Rickards, 2013). Silvestri et al. (2016) studied for the first time the impact of TS on health-related quality

of life of individuals with TS and reported lower health related quality of life with special attention to anxiety, which may be possibly due to the mentioned issues. Moreover, individuals with TS have the constant demand to suppress their tics in social situations, which is reported as being stressful and uncomfortable, and can diminish their social and academic function (Draper, Jackson, Morgan, & Jackson, 2016).

To date there is no cure for TS. Treatments aim to diminish tic severity and frequency. These can be behavioural (mainly awareness training of the PU), pharmacological (dopamine antagonist DA2 are the most used to relieve tic burden) or surgical (deep brain stimulation targeting the thalamus) (Cavanna, Black, Hallett, & Voon, 2017).

1.2.1. Pathophysiology of TS

TS is associated with a dysfunction in the cortical-striatal-thalamic-cortical (CSTC) circuit, although it is important to state that the cerebellum has also been reported to be involved (McCairn, Iriki, & Isoda, 2013). There are three overlapping CSTC circuits: the sensorimotor circuit, the limbic (emotional) circuit and the associative (cognitive) circuit. These circuits are characterized by the fact that direct information from cortical structures to subcortical structures, and back to cortical structures. Subcortical structures include the basal ganglia (BG) nuclei and the thalamus. The BG nuclei, which include the Globus Pallidus internus (GPi) and externus (GPe), Substantia Nigra pars reticulate (SNr) and pars compacta (SNc), subthalamic nucleus (STN), and the striatum (which includes caudate and putamen), comprise motor, associative and limbic territories. The striatum, for example, receives input from the primary motor and sensory motor cortices in the dorsolateral striatum (i.e. motor territory), from the orbitofrontal, cingulate and insular areas in the ventromedial striatum (i.e. limbic territory), and from the prefrontal cortex and association cortices in the dorsomedial striatum (i.e. associative territory). At the end of the loops, the thalamus receives the input from the BG and directs the information back to the cortex.

Although the exact pathophysiology is unclear, it is thought that the cause of the phenotype of the disease is that the striatum exhibits spontaneous firing, leading to the disinhibition of the thalamus and overexcited sensorimotor areas in the cortex, causing tics to be produced. Xu et al. (2015) supported this view showing the key role of the striatum on tic-like phenomenology by creating a rat model for TS showing the typical symptoms of the disease by just disrupting cholinergic interneurons in the striatum. Worbe et al. (2013) also demonstrated tic-like movements in primates by disrupting striatal function. Although the sensorimotor loop is involved more prominently in the execution of tics, it is thought that the associative and limbic loops also play a role in tic execution, as they have shown to correlate with the complexity of the tics and the symptoms of obsessive compulsion, respectively (Worbe et al., 2012).

It is also thought that inhibitory networks are responsible for the disease too, as it is thought that the inability of individuals to inhibit the aberrant overactivity in the BG is a result of a disruption in the inhibitory networks. This disruption has been seen in many TMS studies. A clear example is the study published by Heise et al. (2010), where they examined the intra-cortical inhibition in the M1, which is associated to GABA-A receptors, and found a reduced inhibition in TS individuals at rest compared to controls, and consequently, increased cortical excitability.

Tics can be compared to habit formation in that they are produced without awareness, are repetitive and involve reinforcement learning by rewards (H. H. Yin & Knowlton, 2006), being the reward the relief of the PU. This reinforcement is carried out by dopaminergic neurons (Schultz, 1997) within the SN (Hashemiyoon, Kuhn, & Visser-Vandewalle, 2016). Dopamine networks in TS have been found to be altered (Liao et al., 2016), and for this reason, it is thought that individuals with TS acquire an automatic behaviour more strongly and find it more difficult to unlearn it, becoming a habit (G. Jackson, Draper, Dyke, Ramani, & Jackson, 2015; Delrome et. al, 2016).

On the whole, it is thought that the execution of tics is the consequence of the abnormal spontaneous activity in the striatum leading to an overactivity in cortical motor areas as well as the inability of the subjects to suppress this overactivity. These actions are then reinforced by the feeling of release of the PU leading to the repetition of the behaviour.

1.2.2. Functional and structural connectivity in TS

TS individuals present an increase in the functional connectivity, specifically in the networks between the cortex and the BG (Worbe et al., 2012). This was seen, for example, in J. Wang et al. (2011) study, where they found an increase of bold-oxygen-level dependent (BOLD) responses in the motor cortex (primary motor cortex (M1), supplementary motor area (SMA), premotor cortex (PM), prefrontal cortex (PFC) and somatosensory cortex) and BG (putamen, caudate, pallidum, thalamus, and substantia nigra) in spontaneous tics executed by TS individuals compared to imitated tics executed by healthy individuals, suggesting a stronger functional interaction in the cortico-basal ganglia networks.

TS individuals also present a loss of hubs (Worbe et al., 2012), which leads to widespread disruptions of functional connectivity as seen in anatomically informed computational models (Bullmore & Sporns, 2009). For example, they present a disorganized functional connectivity in the sensorimotor, associative, and limbic CSTC networks (Worbe et al., 2012).

TS individuals also exhibit an increase in the structural connectivity found in an increase in the number of white matter (WM) tracts linking the several motor cortical regions with the striatum and thalamus, which is positively correlated with tic severity (Worbe et al., 2015). The properties of the tracts linking the cortex with the striatum are also altered, presenting an increase of the fractional anisotropy (FA) and a decrease in the radial diffusivity (RD) (Worbe et al., 2015) that are correlated with tic severity. FA is an indicator of axonal density and myelin ensheathment, where an increase in FA is related to an increase in

axonal density and increase in myelin ensheathment. RD is an indicator of myelin density, where an increase in RD is associated to the lack of myelin. Worbe et al. (2015) argued that the results suggest that patients with TS present higher axonal density and demyelination with potential enlargement of myelin sheets.

1.2.3. Neurobiology of premonitory urges in TS

The sensorimotor cortex (SMC) refers to the strongly connected primary motor cortex (M1), located in the pre-central gyrus, and primary somatosensory cortex (S1), located in the post-central gyrus. The main function of the M1 is motor output while the main function of the S1 is to process somatosensory information. In subjects with TS, the activity in the M1 has been associated with tic execution while the activity in the S1 has been associated with PU (Neuner et al., 2014; J. Wang et al., 2011). The function of M1 and S1 is consolidated. Very often, subjects with TS blame their tics on their PU. This was reported in a study of 135 TS participants between 8-71 years old, where 92% declared that their tics were a response to their PU (Leckman et al., 1993). It has been proposed that sensory phenomena, represented in the S1, leads to unwanted actions (S. R. Jackson, 2011).

An activation likelihood estimation (ALE) meta-analytic study reported overlapping areas that activate during the urge to swallow, urinate, yawn and tic, manifesting the similarity between PU and natural urges (S. R. Jackson, 2011). These areas corresponded to the insular cortex and cingulate cortex. A review of studies exploring the neurobiology of PU stated that the insula and cingulate cortex together with the SMA are thought to be the neurobiological signature for PU, as they are activated just before the tic (Cavanna et al., 2017). When the action of the urge is suppressed, it is thought that the insula, which is the area that perceives bodily states, sensors the urge and recruits the ACC in order to suppress the action (S. R. Jackson, 2011; Alicja Lerner et al., 2009). The SMA has been associated with the conscious intention to move (Desmurget & Sirigu, 2009) and therefore, it is likely that the SMA is a mediator

between the urge and the movement execution (Rajagopal, Seri And, & Cavanna, 2013). This suggests that the strong link between PU and tics could be due to a strong connection between the limbic and motor areas.

1.2.4. Tic suppression

Even though tics are usually reported as involuntary actions, many individuals are able to suppress them for a short period of time. This usually leads to an increase in their PU to a point that becomes unbearable resulting in the execution of bouts of tics. Ways of controlling tics involve (I) the conscious effort of inhibiting tics, which can be described by individuals as painful; (II) attentional distraction, to a certain demanding task or physical activity; (III) behavioural training focusing on removing the habit of ticcing to release the feeling of urge (Ganos et al., 2018).

Peterson et al. (1998) were the first to study tic suppression examining the difference of BOLD responses when voluntarily suppressing tics and when ticcing was allowed. They found that the regions involved in tic suppression are the ones implicated in the inhibition of unwanted impulses, which include the prefrontal, parietal, temporal, and cingulate cortices.

It has been suggested that the gain of control of tics is due to the development of compensatory mechanisms that enhance motor control, as it has been reported that individuals with better levels of cognitive control show less ticcing (S. R. Jackson et al., 2011). This is supported by Jung, Jackson, Parkinson, & Jackson (2012), who studied the BOLD responses to a cognitive control of motor output task and showed that patients presenting an increased BOLD response in the M1, SMA, ACC and inferior frontal gyrus (IFG), which are areas involved in response inhibition in impulse action (Q. Wang et al., 2016), were better at the task and showed lower YGTSS scores. Interestingly, these areas have been found to be involved in tic suppression. Zapparoli et al. (2016) found increased activity in the SMA during the imagination of movement and execution of movement, while tics were indirectly being suppressed, and found

a correlation with tic severity; Ganos et al. (2014) found increased activity in the left IFG when TS individuals voluntarily inhibited tics as opposed to free-ticcing examined by resting functional magnetic resonance imaging (fMRI); Peterson et al. (1998) showed increases in the ACC when making the same comparison. These studies support the thought that tic suppression is mediated by areas responsible for cognitive control of unwanted impulses.

Compensatory mechanisms also involve the change in the structural and functional connectivity. S. R. Jackson et al. (2011) found an association between the WM structure in the corpus callosum and tic severity, where individuals with more WM presented more severe tics. An additional behavioural study examining the control of motor output confirmed that this increase in WM is a neuroplastic adaptation to gain control of tics. Additionally, GABA levels are increased in the M1 in TS individuals with the ability to suppress tics, presenting a localized increase in tonic inhibition (Orth, 2009). In fact, a recent paper used TMS to measure cortical excitability in subjects with TS while they were free-ticcing and while they were actively suppressing their tics (Ganos et al., 2018), and showed that subjects were able to suppress their tics by reducing their cortical excitability and the amount of suppression was correlated with the amount of reduction of the cortical excitability. On the other hand, Draper et al. (2014) found increased GABA levels within the SMA as well as decreased activity in that region, when scanning TS individuals, which was inversely correlated with tic severity. Then, increased GABA levels in the M1 and increased GABA levels in the control area of the SMA is related to tic suppression in TS.

Many patients have reported that engaging in complex motor tasks can suppress their tics. Zapparoli et al. (2016) suggest that tics can be reduced when executing a movement or imaging the execution of the same movement due to a competition network with the network responsible for producing the tics. Heise et al. (2010) found an increase in inhibition just before a movement was executed compared to healthy subjects. They interpreted that when the motor cortex was about to perform a voluntary movement, the motor areas and PFC operate as a ‘top-down’ control over the abnormal firing of the subcortical

areas, suppressing the spontaneous activity of the motor cortex and consequent ticcing. Zapparoli et al. (2016) supported this model with the finding that the PM and prefrontal areas are hyper-activated during the imagination of movement and execution of movement and tics are suppressed.

Taken together, tics are the result of excessive activity in the motor areas in the cortex, as well as poor cognitive control to inhibit the motor output. PU are the result of excessive activity in the S1 and limbic areas, and it is thought that the strong connection between S1 and M1, as well as the strong connection between limbic areas and motor areas are the cause of the strong link between PU and tics**.** Suppression of tics is achieved by reducing the excessive activity in the motor cortex, likely achieved by cognitive control to inhibit the motor output. Functional and structural connectivity do also play a role in both the generation of tics and the suppression of tics.

1.3. Entrainment of brain oscillations

Brain oscillations reflect the current flow of the neural populations, cycling rhythmically at a specific frequency. There are different well-established rhythms of brain oscillations. These rhythms differ in their frequency, but also in their origin and reactivity to changes in sensory input and task demands. Brain oscillations from each frequency band are thought to play a role in different cognitive functions and neural processing. Alpha/Mu (8-13Hz) and beta (13-30Hz) rhythms are the strongest electrophysiological signals in the SMC. Mu rhythm is associated with the processing of sensory information and beta rhythm is associated with motor control; importantly, both rhythms de-synchronise during movement. Due to the strong association of these rhythms with the SMC and the relation between mu and urges and beta and tic execution, I will be focusing on these two rhythms in the studies presented in this thesis.

The desynchronization of alpha and beta rhythms within a neural population, due to cells firing independently, is thought to facilitate to encode information within a network and is associated with an increased capacity of processing; while a synchronization of alpha and beta rhythms within a neural population, due to cells firing in synchrony, is thought to incapacitate to encode information within a network and is associated with brain lower capacity of processing (Brookes et al., 2015). The measure of synchronization or desynchronization gives a measure of cortical activity.

Cortical activity can be modulated by the application of an external rhythmic stimulation at a specific frequency for a short period of time. This rhythmic stimulation leads to a synchronization of the intrinsic neural oscillations with that external stimulation. When the external rhythmic stimulation interacts with the ongoing brain rhythms, and therefore, modulating brain oscillations, it reflects entrainment; when the external rhythmic stimulation simply overlays evoked potentials, it reflects steady-state evoked potentials.

The successful achievement of entrainment of neural oscillations depends on the duration of stimulation or the number of pulses applied (the effect is stronger when the length of stimulation is longer or the number of pulses is higher), and the frequency of stimulation (the effect is stronger when the frequency of stimulation is closer to the preferred frequency of the neural population being stimulated).

Rhythmic stimulation can be applied with (I) repeated short stimuli delivered at regular intervals (e.g. visual/auditory input, TMS pulses) and (II) oscillatory stimulation delivered at a continuous rate (e.g. transcranial alternating current stimulation (tACS)). Published entrainment effects have been acquired using sensory input, repetitive TMS (rTMS), and tACS; being rTMS and tACS the most used techniques. rTMS involves firing of the neurons and thus, evokes entrainment by resetting of the phase of the intrinsic oscillations of these cells; on the other side, tACS shifts the neurons' resting membrane potential leading to a change in the neurons' firing rate.

1.3.1. Entrainment effects on behaviour

Provided that brain oscillations play a role in cognitive functions, entrainment has an impact in behavioural performance.

Very few published studies have aimed to entrain the motor cortex to examine behavioural effects, and the ones that have, have focused on entraining the beta band. Alpha-entrainment has only been used so far to examine its effects on the occipital cortex and visual perception. Here I review the papers entraining the motor cortex and looking at its effects on behaviour.

Joundi et al. (2012) entrained beta band in the motor cortex at 20Hz using tACS during a go/nogo task in which participants had to respond by squeezing a grip force sensor. Results showed that entrainment reduced the rate of force generation in both the go and no-go condition. Pogosyan et al. (2009) entrained beta band in the motor cortex at 20Hz using tACS during a voluntary movement task in which participants had to track a circle using a joystick. Results showed that entrainment reduced the velocity of the voluntary movement. Brittain et al. (2013) was able to reduce tremor in 5 subjects with Parkinson Disease by entraining the motor cortex at the preferred tremor frequency at the optimal phase for tremor suppression using tACS.

Although these studies did not differentiate between entrainment and steady-state evoked potentials, the fact that the stimulation had an effect on motor behaviour suggests that they were able to modulate brain oscillations through entrainment.

1.3.2. Aftereffects of entrainment

Effects of entrainment persist after the stimulation although phase entrainment only lasts a few oscillations. A review from 2015 of papers examining both oscillatory and behavioural aftereffects (Veniero, Vossen, Gross, & Thut, 2015) showed that from 22 studies examining aftereffects of entrainment using

tACS/tDCS, 19 reported aftereffects; and from 26 studies examining aftereffects of entrainment using rTMS, 22 reported aftereffects. They also reported that these effects last between a few minutes to half an hour for tDCS and 65 min for rTMS.

A relation between the strength of entrainment and the strength of the aftereffects has been shown (Helfrich et al., 2014) suggesting that aftereffects have a mechanism dependent from entrainment. Coherent with this, it has been suggested that the aftereffects are due to the strengthening or weakening of neuronal networks by spike-timing dependent plasticity lead by entrainment of the brain oscillations (Veniero et al., 2015).

1.4. Summary and research aim

TS is characterized by the execution of involuntary, repetitive, stereotyped behaviours called tics. Most individuals report having uncomfortable sensations preceding a tic called premonitory urges, and many individuals report that their tics are a result from these sensations. Tic execution is associated with increased excitability in the motor cortex and premonitory urges are associated with an increased excitability in the somatosensory cortex. Tic suppression can be achieved with voluntary movement and is associated with a reduced excitability in the SMC. An increase in the synchronization of brain oscillations is associated with a reduction in cortical excitability. It is possible to increase the synchronization of brain oscillations with entrainment. Provided that brain oscillations play a role in cognitive functions, entrainment of the SMC will have an effect on motor behaviour.

This thesis has two main aims: (I) to explore the brain activity preceding voluntary movements and preceding tics, and (II) to investigate the use of a non-invasive brain stimulation technique to modulate and inhibit this brain activity in order to generate an effect on motor behaviour including voluntary movements and tics.

CHAPTER 2

Chapter 2 - General methods

2.1. Participants and clinical measurements

2.1.1. TS Participants

All the patients who took part in the studies had a confirmed diagnosis for TS. I declare that data presented in Chapter 3a.1, 3a2, 3b, 4, and 6 was collected by myself, and that data presented in Chapter 5 was collected by two undergraduate students supervised by our research team.

In all studies, participants were asked to read the information sheet carefully; further explanation of the used technique (EEG/fMRI/MNS) was offered, and any questions answered. The participants' right to withdraw at any time point was emphasized. Participants or their parents or guardians, depending on whether the participant was over 18 or under 18, respectively, were asked to fill in the consent form. In the study using MRI, participants or their parents or guardians were also asked to complete an MRI safety questionnaire. At the end of the experiment, all participants were debriefed. All information was anonymized, stored securely under a locker and only used for the purpose of the study. All studies were approved by the Ethics Committee in Nottingham University.

Patients under 18 were screened for symptoms of Attention-deficit hyperactivity disorder (ADHD) using the Conners 3-Parent measure (Conners, 2008), obsessive-compulsive behaviour (OCB) using the Child YGTSS-Brown Obsessive-Compulsive Scale (CY-BOCS; Scahill et al., 1997) rating scales, emotional and behavioural problems using the Strengths and Difficulties Questionnaire (SDQ; Goodman, 1997), and autism spectrum disorder (ASD) using Social Communication Questionnaire (SCQ; Berument, Rutter, Lord,

Pickles, & Bailey, 1999). Adult participants were screened for symptoms of OCB using YGTSS-Brown Obsessive-Compulsive Scale (Y-BOCS; Goodman, Price, Rasmussen, Mazure, Delgado, et al., 1989) and ASD using Autism Spectrum Quotient (AQ; Baron-Cohen, Wheelwright, Skinner, Martin, & Clubley, 2001). The IQ score was estimated from all participants whom we did not have previous records from using the Wechsler Abbreviated Scale of Intelligence - II (WASI-II; Wechsler, 1999) test. Patients were also asked whether they had been diagnosed with other psychiatric disorders, and all current and past medication taken by participants and years of education in adult participants were recorded. The only exclusion criterion was IQ below 70.

2.1.2. Yale Global Tic Severity Scale (YGTSS)

Yale Global Tic Severity Scale (YGTSS; Leckman et al., 1989) is a semi-structured clinician-rated scale of tic severity over the past week. The measure involves the rating of motor-tic severity, vocal-tic severity, and tic-related impairment severity. The rating of motor- and vocal-tic severity is made by assessing the number, frequency, intensity, complexity and interference of the motor and vocal tics separately; each of these rated in a scale from 0-5. Therefore, motor-tic severity and vocal-tic severity measures have a maximum possible score of 25 each. The tic impairment is assessed by asking the patients to rate the distress and impairment of having tics experienced in the daily life in the interpersonal, academic, and occupational environment. The rating of the impairment is made in a scale from 0 to 50, where 0 indicates that having tics does not cause any distress to the patient, and 50 indicates that having tics does cause a lot of distress to the patient. Overall, the tic-severity measure has a minimum possible score of 0 and a maximum possible score of 100.

Reliability and validity for this scale have been supported in a study examining the internal consistency and stability of its scores (Storch et al., 2005).

2.1.3. Premonitory Urge for Tics Scale (PUTS)

PUTS (Woods, Piacentini, Himle, & Chang, 2005) is a psychometric measure of tic-related premonitory urge. The original version (Woods et. al, 2005) consists of 10 questions rated from 1 (“Not at all”) to 4 (“Very much”) assessing the type of sensory feeling, its frequency and the relationship between the premonitory sensory phenomena and the tic. The total score is calculated by adding the score of 9 of these 10 items, being 9 the minimum possible score and 36 the maximum possible score.

The assessed PUTS used in the studies presented in this thesis corresponds to a new version of PUTS scale that has recently been published (Baumung et al., 2020). This new version, known as Premonitory Urge for Tics Scale – Revised (PUTS-R), has 8 of the 9 scored items from the original PUTS plus additional questions. Overall, the test consists of 24 questions rated from 0 (“Not at all”) to 4 (“Very much”). Fifteen of the questions refer to how these feelings feel like (e.g. “Right before I do a tic, I feel like there is energy in my body” or “Right before I do a tic, I have a feeling that is similar to an ache in my head or body”), five of the questions refer to how often these feelings precede tics (e.g. “I have these feelings almost all the time before I do a tic” or “I rarely have these feelings before a tic”), and four of the questions refer to how strong these feelings are overall (e.g. “These feelings are strong” or “This feeling gets stronger until I do a tic”). Since there is no set scoring system for PUTS-R yet, the total score was calculated by adding the score of the 8 items also scored in the original PUTS, being 0 the minimum possible score and 32 the maximum possible score.

It has been pointed out that PUTS measures are limited, as it considers all PU in a single scale, and therefore specific PU are not distinguished for different tics (Valerie Cathérine Brandt, Beck, Sajin, Anders, & Münchau, 2016)), which prevents the measure of PU in different subscales. However, reliability of this scale has been shown for youth older than 10 years of age (Woods et al, 2005), demonstrating good internal consistency and temporal stability (Reese et al., 2014; Woods et al., 2005); and validity of this scale has been shown in a study

examining the correlation between PUTS score and a real-time urge measure based on the intensity and frequency of urges (Valerie Cathérine Brandt et al., 2016). PUTS score has also been shown to be correlated with the total score of the YGTSS (Reese et al., 2014; Woods et al., 2005).

2.2. Electroencephalography (EEG)

EEG is a non-invasive functional brain imaging technique that uses electrodes placed on the scalp to record electric events taking place in the brain. These events can be either changes in electric potentials or changes in the electric fields generated by the electric activity from the active pyramidal neurons in the cortex. EEG can quantify the magnitude of these electric events as a function of time and frequency, which is usually referred as EEG power.

In this chapter, I will briefly explain the neurophysiological basis of EEG, highlight its big limitation and provide an overview of the EEG recording, EEG pre-processing and EEG data analyses.

2.2.1. Neurophysiological basis of EEG

The changes in electric field recorded by EEG correspond to the changes in extracellular potentials produced by the sum of excitatory and inhibitory postsynaptic potentials of the pyramidal cells in the cortex (Kirschstein & Kohling, 2009). EEG is able to measure the voltage of these potentials in time.

Brain oscillations are generated by the synchronized extracellular potentials of connected neurons in neural populations and the time for the signal to travel from the dendrites through the axon to the synaptic button and released (Kirschstein & Kohling, 2009). Brain oscillations can be found all over the brain and in different frequencies ranging from approximately 0.05Hz to 500Hz (Staba, Wilson, Bragin, & Fried, 2002). EEG is able to measure the magnitude and synchronization in phase of these brain oscillations.

2.2.2. Limitation: Spatial resolution

EEG has a very high temporal resolution but low spatial resolution. The reason of this is volume conductivity (Wendel et al., 2009). The layers in the brain (muscle, cerebrospinal fluid, grey and white matter) as well as the scalp and skull have different conductivities, capacity of forming an electric field (i.e. permittivity), and magnetic permeabilities, and because of that, the signal recorded by each scalp electrode contains a mixed signal coming from different sources, which leads to the commonly known as forward problem. The forward problem is due to calculating the electrode potentials and magnetic field assuming a source distribution inside the brain and conductivity distribution through the head volume. It is reduced by using realistic brain models, which estimate the geometry and conductivity of cortex, CSF, skull, and skin. One example is the *Besa 4-shell* spherical model. However, realistic brain models do not take into account individual's electrical-conductivity distribution. The best solution is to co-register the MRI scan of the subject with the EEG data. The inverse problem, also called neural source imaging problem, comes from the estimation of the position of the current sources from the electrode potentials, because any topography could have been the result of an infinite number of possible sources. The inverse problem is solved by solving the forward problem. Since I did not have individuals' MRI scans from the participants in the EEG studies carried out, I did not perform source localization on the data.

2.2.3. Considerations for high-quality EEG recordings

Different factors should be taken into account when collecting EEG data in order to maximize the quality of our raw data.

The EEG signal recorded by the scalp electrodes includes brain activity as well as non-brain activity (e.g. eye movements and jaw clenches). The use of a reference electrode recording non-brain activity is desired in order to subtract it from the overall recorded EEG signal. It is common procedure to use the left

and right mastoids as reference electrodes (Light et al., 2010). It is also common procedure to monitor for eye movements, as these belong to the non-brain activity signal recorded by the EEG electrodes (Light et al., 2010). In order to do this, four electromyography (EMG) electrodes are placed around the eyes, two vertical EOG electrodes and two horizontal EOG electrodes that will monitor vertical and horizontal eye movement, respectively.

It is important to keep electrical impedances low in all EEG and EMG electrodes, since high impedances distort the EEG signal. For this reason, it is common to ask participants to come with no hair gel or make-up on the day of the testing, as these could easily distort the EEG and EMG signal, respectively, and provide them with skin cleansing swabs to clean the regions of the face were the EMG electrodes will be placed (Light et al., 2010). It is also common to use an electroconductive gel to create a bridge between the skin and the electrode which reduces electrical impedance (Light et al., 2010). It is usually accepted to keep impedances below 5 kΩ (Light et al., 2010).

The use of a high number of electrodes increases source localization accuracy, however, it also increases the time spent on set-up, which can be undesirable when testing children or patients. It is common to use a number of 64 electrodes and to measure the head of participants with a measuring tape in order to use an electrode cap of the right size (Light et al., 2010). The most common placement of the electrodes is the international 10-20 system (Jasper et. al, 1958). This system places the electrodes based on skull landmarks and covers all the head.

EEG data can be recorded in different sampling rates. The use of a high sampling rate, increases temporal resolution and increases the frequency that is captured.

Participants are usually placed in a Faraday cage when recording their brain activity, making sure that they do not carry any electronic objects (e.g. smart

watch, wallet) with them. The Faraday cage ensures that external electromagnetic fields are not picked up by the EEG electrodes.

2.2.4. Experimental set-up used in the presented studies

In all the studies in this thesis that used EEG, participants were asked to come with no hair gel or make-up on the day of the testing. They were given a skin cleansing swab and they were asked to clean the regions of the face where the EMG electrodes were going to be placed. A total of 4 EMG electrodes were placed on the face: two vertical (V)EOG electrodes, two horizontal (H)EOG electrodes, and the reference electrodes were placed on the left (M1) and right (M2) mastoids. The head of each participant was measured with a measuring tape and an electrode cap of the right size was adjusted to each participant's head. The number of electrodes used was 64 and the placement of the electrodes used was the international 10-20 system. Figure 2.1 shows the position of the electrodes on the scalp. Each electrode hole was filled with salt-based gel (*SignaGel; Parker Laboratories, Inc.*) with a blunt syringe to obtain a good signal. When necessary, hair was rubbed to ensure gel was applied on the scalp. Electrodes were then attached to the cap. When the set up was finalized, participants were placed in a Faraday cage (2.5 x 2.0 x 2.3 m) to minimize external electromagnetic fields. I ensured that all electrodes had the right impedance. After the experiment, participants were offered to remove the gel from their hair with shampoo and water.

EEG data was recoded reference-free from 64 electrodes using BioSemi Active Two System. EEG data was recorded with a sampling rate of 1024Hz. While in the Faraday cage, participants were monitored via a web camera (Logitech C920 PRO HD Webcam). Lights from the Faraday cage were turned off while recording, but a LED light connected to a thunderbolt dock inside the cage was on, so that participants were not in the dark during the testing. This was done to avoid noise at 50Hz.

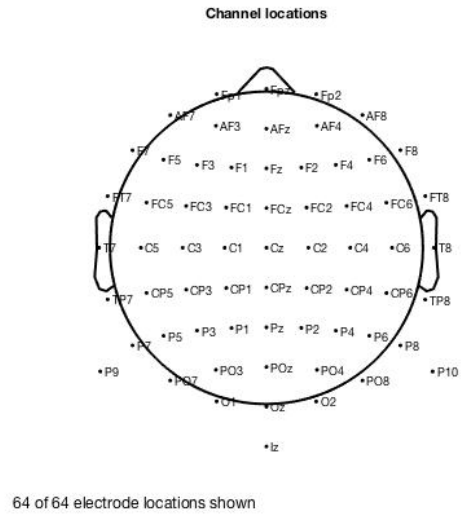


Figure 2.1. Channel locations with the name of the electrodes

2.2.5. Pre-processing steps of the EEG data

Main references used in this section (2.2.5) are EEGLAB tutorial (Delorme, Fernsler, Serby, & Makeig, 2006); and Makoto's preprocessing pipeline by the *Swartz Center for Computational Neuroscience (SCCN)* (Makoto's preprocessing pipeline. (n.d.). Retrieved April 23, 2020).

Even though the amount of noise collected during EEG recording is minimized, there will still be noise in the signal. For this reason, it is common to follow some pre-processing steps with the main aim to reduce the noise in the data without reducing the amount of signal. At this point the EEG electrodes are also called channels.

Pre-processing steps involve downsampling the data in order to reduce computational time and disk space; low-pass filtering removes high frequency noise and high-pass filtering removes low frequency noise; applying a realistic brain model that estimates the geometry and conductivity of the brain; subtracting the signal from the reference electrodes from the data; delete or interpolate bad channels, interpolation uses the surrounding channels to make an educated guess of the activity at the bad channel and is done to keep the

number of dimensions of the EEG data; removal of eye blinks and eye movements from the data; baselining, which is used to relativize the data of interest to a resting period.

Usually, EEG data is used to examine the brain activity associated with an event. In order to do that time-windows time-locked to the event, known as epochs, are extracted and those epochs showing excessive noise are deleted. Usually those epochs showing a signal amplitude at +-100μV in one or more channels are rejected, those epochs with signal slopes exceeding a threshold of 50μV in one or more channels are rejected, those epochs with 5 times the standard deviation in the probability distribution are rejected; those epochs which their kurtosis statistic larger than 5 times of the standard deviation of the data are rejected.

Independent Components Analysis (ICA) identifies those components that show spatially and temporally related brain activity. Thanks to that, it is possible to get rid of activity that is not brain-related (e.g. blinks, saccades and muscular contractions that have not been removed in previous pre-processing steps). When a channel has been deleted, and therefore, there is a reduction in the number of dimensions, it is standard procedure to run a Principal Component Analysis (PCA) before running the ICA to reduce the number of dimensions to the ones present in the data. This is done to minimize the computational time and the amount of data that is required for the ICA. This step has no implications for the resulting number of ICA components.

2.2.6. Pre-processing used in the presented studies

EEGLAB 14 (14.1.1) was used to pre-process the EEG data.

Data was down sampled to 128 Hz. Then, data was low-pass filtered, and high-pass filtered at the desired frequency depending on the aim of the later analyses: when examining the mu band, I filtered from 8-13Hz; when examining the beta band, I filtered from 13-30Hz; when examining the event-related

potential, I filtered from 1-45Hz. Since I did not have the MRI scan of the participants, channel locations were applied using Besa 4-shell spherical model, which estimates the geometry and conductivity of cortex, CSF, skull, and skin. Brain channels were referenced to the linked mastoids. By doing this, the average signal of the two mastoid channels was subtracted from every channel in the scalp. After that, bad channels were visually identified. Those channels showing aberrant behaviour were deleted, those channels showing excessive noise were interpolated. When a bad channel was deleted or interpolated, data was re-referenced to the linked mastoids. Automatic Artifact Removal (AAR, view Gómez-Herrero, G. (2007) for info) was used to remove EOG artefacts using recursive least squares regression. This algorithm has been shown to remove blinks and saccades without altering the underlying EEG data. After that, I subtracted the z-scores to bring the mean of the data to 0 and standard deviation to 1 in order to standardize the data. Because of this step, no baselining of the data was needed.

Then, I extracted the time-windows time-locked to the event of interest. Epochs were then rejected: those epochs showing a signal amplitude at +-100μV in one or more channels were rejected; those epochs with signal slopes exceeding a threshold of 50μV in one or more channels were rejected; those epochs with 5 times the standard deviation in the probability distribution were rejected; those epochs which their kurtosis statistic was larger than 5 times of the standard deviation of the data were rejected.

After the epoch rejection, ICA was run. If a channel had been deleted, and therefore, there was a reduction in the initial number of dimensions, I ran a Principal Component Analysis (PCA) before running the ICA to reduce the initial number of dimensions (i.e. 64) to the ones now present in the data (i.e. 64 minus the number of deleted channels). Components were rejected with the use of Multiple Artifact Rejection Algorithm (MARA). MARA is a tool that uses expert ratings to automatically identify artefactual independent components by classifying them based on the extraction of six features from the spatial, the spectral and the temporal domain. ICA components were also rejected by own visual inspection.

2.2.7. Analysis of the EEG data

The collected EEG data allow us to examine the changes in evoked potentials related to a specific event (event-related potentials or ERP) and the changes in the electric fields (event-related spectral perturbation (ERSP)) compared to the baseline. ERPs show the synchronized firing of pyramidal neurons due to a specific stimulus and offers information about the timing and location (based on the highest amplitude of the potential) of neural events involved in cognitive processes. ERSP shows the changes in the synchronization of fluctuations of neuronal populations oscillating at a specific frequency, due to a specific stimulus, and offers information about the state (activated/inhibited) of the cortex.

When there is an increase in synchronization in the brain oscillations of a neural population, we talk about event-related synchronization (ERS), while when there is a decrease in synchronization in the brain oscillations of a neural population, we talk about event-related desynchronization (ERD). ERS can be evoked by either an increase in amplitude of the brain oscillations, an increase in phase synchronization of the brain oscillations, or both (Kirschstein & Kohling, 2009). On the other side, ERD can be evoked by either a decrease in amplitude of the brain oscillations, a decrease in phase synchronization of the brain oscillations, or both (Kirschstein & Kohling, 2009). These event-related rhythms can be measured with the increase (related to synchronization) or decrease (related to desynchronization) in power of the frequency band of those neural populations, in relation to the baseline.

Inter-trial coherence (ITC) analyses quantify the consistency in which oscillations fluctuate at the same phase and rhythm across trials and is used as a measure of phase alignment or phase synchrony. Values of ITC can also be referred as phase-locking values (PLV).

Time-frequency (TF) analyses are used when computing ERSP analyses and ITC analyses of the event-related rhythms. Fast Fourier Transform (FFT) is a common algorithm used for TF analyses and is the one implemented in the work presented here. While ERP looks at changing voltage over time, FFT indicates what voltages are present in each frequency in the data.

EEG data analyses can also be performed to cluster the ICs of the subjects in order to examine scalp topographies. Usually, only IC with less than 15% residual variance are used for the group analysis (Artoni et al., 2014). Clusters are based on the spatial homogeneity of functionally similar or equivalent IC processes across subjects (Julie Onton & Scott Makeig, 2006).

2.3. Magnetic Resonance Imaging (MRI)

2.3.1. Introduction

The MRI relies on the properties of nuclear magnetic resonance. When applying a strong constant magnetic field, molecules resonate at a specific frequency producing an electromagnetic signal that can be measured by radio frequency coils (Jezzard & Clare, 2001).

The MRI study presented in this thesis was collected in the Sir Peter Mansfield Imaging Centre at the University of Nottingham. The aim of this study was to detect changes in BOLD signal throughout the brain in an event-related design, therefore, in this section I am going to focus on explaining the general methodology for hemodynamic response detection and whole-brain analyses.

The book *Handbook of functional MRI data analysis* (Poldrack, Mumford, & Nichols, 2011) has been used as reference for the description of fMRI processing and fMRI data analysis presented in this section (2.3).

2.3.2. Structural MRI

The structural MRI scan is a high-resolution anatomical image of the brain with a clear contrast between the white matter and the grey matter. This is captured by the T1-weighted image. This scan is performed to every subject in order to have a detailed image of their brain, which the functional image does not provide.

2.3.3. Functional magnetic resonance imaging (fMRI)

The neuronal activity induced by a stimulus or a task results in an increase in oxygenated haemoglobin in the areas of the brain involved in that activity (this is referred as a hemodynamic response). This results in an increase of the magnetic susceptibility, which can be detected by an MRI scanner. We refer to this signal as Blood Oxygen Level Dependent (BOLD) signal. In fMRI, we want to detect the hemodynamic response, by measuring the BOLD signal, and locate it in the brain. This is captured by the T2*-weighted fMRI image.

The Hemodynamic response function (HRF) is characterized by an initial dip, which is thought to reflect the initial oxygen consumption that happens just before the BOLD changes. This initial dip is followed by a positive peak. The height of the peak depends on the amount of activity present in the tissue, the time of peak is usually between 4-6 seconds after the stimulus onset, and the peak width lasts between 1-2 seconds. After the peak, there is a post-stimulus undershoot. Overall the HRF signal takes 12-20 seconds to return to baseline. See Figure 2.2.

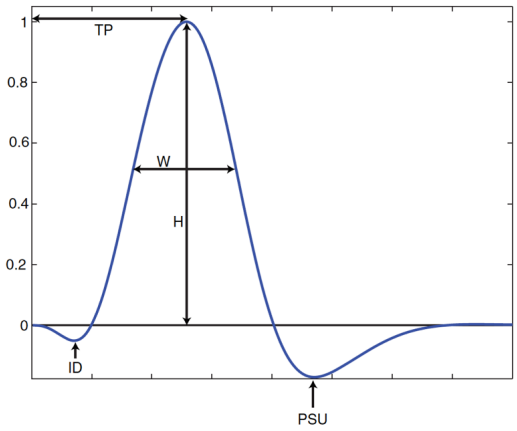


Figure 2.2 Shape of the HRF. ID: initial dip; TP: Time of the stimulus until peak; H: height of response; W: width of the HRF at half the height; PSU: post-stimulus undershoot. This image has been taken from The Handbook of functional MRI Data analysis.

The General Linear Model (GLM) is used to fit and detect BOLD signal. This approach relies on the assumption of the linearity between the neural response and the BOLD signal, which assumes that when a neuronal response is increased, the BOLD response is also increased by the same amount, and that when the stimulus is shifted in time, the BOLD response is shifted by the same amount of time.

In order to detect the BOLD signal using GLM, the HRF is first estimated by averaging all the evoked responses to the same stimulus (this is referred as deconvolution). Then, the BOLD signal is predicted by passing the HRF over the averaged signal, resulting in an added response at each point where both function and signal overlap (this is referred as convolution). Usually, the function used for the HRF is a combination of two gamma functions, where the first gamma function models the initial BOLD response corresponding to the initial dip and positive peak, and the second gamma function models the post-stimulus undershoot.

In order to best predict the BOLD response to a specific stimulus, a statistical model incorporating different parameters to the GLM is fitted. This allows for a bigger flexibility of the shape of the HRF, and thus a bigger variability in the estimates of the HRF. Common procedure is to include motion parameters plus

derivatives, which take into account any small-time offsets of the head displacement, as well as temporal derivatives, which take into account any small-time offsets of the peak of the response, and include them as nuisance regressors to the statistical model.

The estimation of the BOLD response is done in three levels: the single-run analyses (first level), were all the responses of interest to the same stimulus are averaged within runs; single-subject analyses (second level), were all the first-level effects are averaged between runs; and group analyses (third-level) were second-level effects are averaged between subjects.

In first-level analyses, brain images will need to be aligned using image registration techniques. First, the fMRI brain images of each subject will be aligned to their corresponding anatomical brain. This is referred as co-registration. Then, this co-registered image will be aligned to a common spatial framework using a template. This is referred as normalization. See Figure 2.3. The most common used template is MNI152, which is a T1-weighted average structural image corresponding to the average of 152 adult healthy brains. The aim of normalization is to be able to combine the data of all the subjects to the same space for group analyses (i.e. third-level).

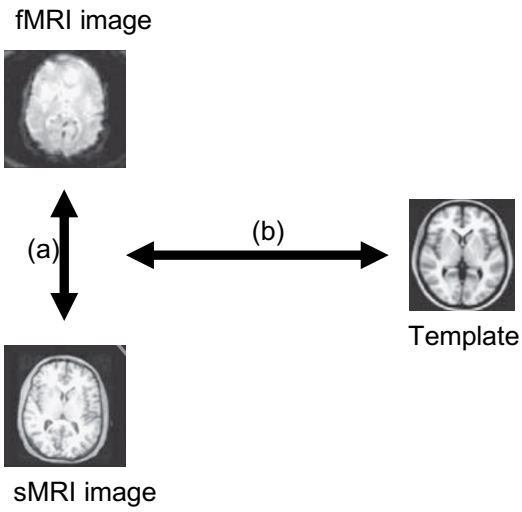


Figure 2.3. Scheme showing the process of MRI image registration

Image registration techniques use the information of each voxel (3D pixels) of the images to be aligned. Image registration is done in 4 steps: first, a cost function that will measure the difference between the two images that we want to align is used; then, the parameters that will be used to transform the image to be aligned are estimated; once we know which parameters will be used for the transformation, these parameters are applied to the image to be aligned; finally, the new voxels have to be interpolated with the original ones in order to obtain the resliced image.

Once the BOLD response has been estimated, the statistical significance of the signal is evaluated. The most common way of doing this is by finding groups of connected voxels that show activation and testing the significance of that group (this is referred as 'cluster-level' inference). The cluster-forming threshold is defined by a specified z-threshold of the z-statistic map. It is usually not acceptable to use a z-threshold lower than 3.1. Gaussian Random Field Theory (RFT) is used to find those clusters over the threshold that have a P value of 0.05 or lower and family-wise error is used to correct for multiple comparisons.

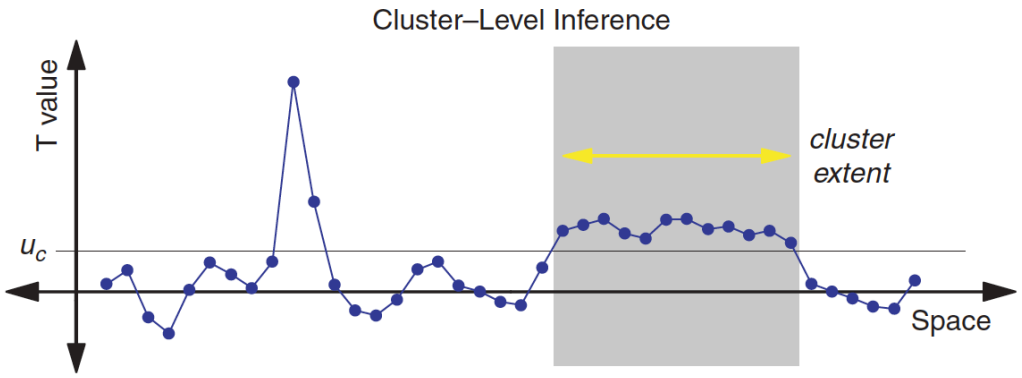


Figure 2.4. Illustration of cluster-level inference. Those clusters over the cluster-forming threshold (uc) are found significant. This image has been taken from The Handbook of functional MRI Data analysis.

Once we have the significant BOLD increase or significant BOLD decrease, we need to locate it in the brain. In order to do that, probabilistic brain maps are the most commonly used. These brain maps give a percentage of probability that the specified voxel belongs to a certain brain structure. The brain maps used for the study presented in this thesis are Harvard-Oxford Subcortical Structural

Atlas and Harvard-Oxford Cortical Structural Atlas to determine the corresponding structural area, and Juelich Histological Atlas to determine the corresponding Brodmann's area.

2.3.4. Pre-processing of fMRI images

The pre-processing of the fMRI data involves motion correction, brain extraction, spatial smoothing, grand-mean intensity normalisation, high-pass temporal filtering, and pre-whitening.

Motion correction is used to correct for head movement during the fMRI runs. This is done by aligning all the images in the fMRI time series of a run to a common reference image. It is common procedure to use the middle image of the scan as the reference image for motion correction. This is because the middle image is the most similar brain image to the first and last scanned images of the whole run and so the overall transformation cost is minimal. The parameters used to align the images include the three translation parameters, which describe any displacements along the X, Y, and Z axes; and the three rotation parameters, which describe any displacement of pitch, yaw, and roll. These parameters are always used in within-subject registration, where no change in shape is expected. FSL uses MCFLIRT (Jenkinson, 2002) to correct for motion, using normalized correlation ratio as cost function.

Spatial smoothing reduces the noise by removing high-frequency information, which results in the blurring of the image. The common procedure is to convolve the 3D image with a 3D Gaussian filter or kernel, and the amount of smoothing is determined by the width of the distribution (what in statistics is described by the standard deviation, in image processing it is described by the full width at half-maximum (or FWHM)). Common procedure is to use twice the voxel size as FWHM. This step is crucial as it is necessary for the RFT.

Temporal high-pass filtering is done to remove low-frequency noise from the data in all timepoints.

Prewhitening removes any temporal autocorrelation in the data. This is done because a temporal autocorrelation in the data would bias the GLM. In order so that the removed correlation is part of the noise and not part of the signal of interest, the GLM is first fit to the data, ignoring any temporal correlation, resulting in the acquirement of the model residuals (the original data with no modelled variability). These residuals are then used to estimate the voxel correlation structure. After that, prewhitening is applied to both the data and the design matrix. Finally, the model estimation is carried out.

2.3.5. How to deal with high motion in the fMRI data

Even though many subjects are able to stay very still in the scanner, they can't avoid some small movements that evoke an increase or decrease in the intensity of the signal in the brain that can lead to a later misrepresentation of the data. Because of that, it is standard practice to detect those movements in order to either remove the corresponding data points or discard the whole subject depending on how big and/or often the movements are. This is done by estimating the head position in space at each time point and measuring the difference between one image to the next one.

Framewise displacement (FD)

It calculates the difference in the sum of absolute values used to estimate the realignment from one volume to the next one at every timepoint (Power, Barnes, Snyder, Schlaggar, & Petersen, 2012). By convention, the first volume of the run has an FD of 0. Those timepoints with FD values over a specified threshold are then used to create a confound matrix (using FSL Motion Outliers in FSL), that will be used in the GLM model to remove the effects of the selected timepoints from the analyses. Common procedure is to discard those volumes with an FD over 0.9mm.

Absolute mean displacement

It is calculated by averaging the sum of the absolute values of translation and the sum of the absolute values of rotation between two images, usually between the middle image and the volume at each timepoint of the run. By doing this, this measure indexes head position throughout the run, which is then used to discard runs or subjects. Common procedure is to discard a run when the absolute mean displacement is bigger than half a voxel size. This is calculated when performing motion correction using MCFLIRT in FSL.

2.4. Median nerve stimulation (MNS)

Median nerve stimulation is a non-invasive brain stimulation technique that, instead of targeting the brain like TMS or tACS, targets a peripheral nerve. In this chapter, I will explain the anatomy of the median nerve, the effects of MNS in the brain by means of ERP and ERSP, and its different current uses in research.

2.4.1. Median nerve

The median nerve originates in the brachial plexus and innervates the thenar muscles, palmar surface and fingertips of the thumb, index and middle fingers (see Figure 2.5). The brachial plexus connects with the 4 cervical nerves (C5-C8) and thoracic nerve (T1) in the spinal cord, also known as spinal nerves. These spinal nerves (C5-C8 and T1) transmit information from the motor cortex to the shoulder, deltoid, biceps, arm, wrist, hand, intercostals and trunk above the waist.



Figure 5.6. Image showing the course of the median nerve from the finger tips to the brachial plexus. This figure has been taken from the website teachmeanatomy

2.4.2. Median nerve stimulation

An electric pulse on the median nerve, strong enough to evoke a thumb twitch, has been shown to produce an effect in the SMC.

Somatosensory evoked potential (SEP)

The stimulation of the median nerve at an intensity strong enough to evoke a thumb twitch produces somatosensory evoked potential (SEP) components that can be seen in the contralateral SMC, being located in the precentral (i.e. motor cortex area) and postcentral (i.e. primary somatosensory cortex area) gyrus (Truett Allison, Mccarthy, & Wood, 1992). The first SEP component in the postcentral gyrus is found 17-22ms after the pulse (N20) (T Allison et al., 1989; Balzamo, Marquis, Chauvel, & Régis, 2004; Fukuda et al., 2008), and the first SEP component in the precentral gyrus is found 24-26ms after the pulse (N24) (Balzamo et al., 2004).

Effects of MNS in the ERSP

The stimulation of the median nerve at an intensity strong enough to evoke a thumb twitch produces a beta ERD (Hirata et al., 2002; Mullinger, Mayhew, Bagshaw, Bowtell, & Francis, 2014) and mu ERD (Hirata et al., 2002; Mullinger et al., 2014; Nikouline et al., 2000) in the bilateral SMC, but more prominent in the contralateral cortex (Hirata et al., 2002; Mullinger et al., 2014; Nikouline et al., 2000). This is followed by a posterior beta ERS (Chen, Yaseen, Cohen, & Hallett, 1998; Della Penna et al., 2004; Neuper & Pfurtscheller, 2001b; G. Pfurtscheller, Woertz, Müller, Wriessnegger, & Pfurtscheller, 2002) and mu ERS (Della Penna et al., 2004) in the bilateral SMC, but greater in the contralateral hemisphere (Della Penna et al., 2004; Gaetz, Edgar, Wang, & Roberts, 2011; Neuper & Pfurtscheller, 2001b; Salenius, Schnitzler, Salmelin, Jousmäki, & Hari, 1997).

ERD is thought to reflect cortical activation due to the processing of the somatosensory information (Neuper & Pfurtscheller, 2001a), while ERS is thought to reflect cortical inhibition due to the recovery of the oscillatory activity that has been activated to process information.

Different current uses of MNS in research

Median nerve stimulation is an effective tool to:

(I) Examine the activation of different brain areas due to MNS by evaluation of the SEP (T Allison et al., 1989; Truett Allison et al., 1992; Truett Allison, McCarthy, Wood, & Jones, 1991)

(II) Examine the propagation of activity following the MNS from the primary S1 to the secondary somatosensory areas (S2) by evaluation of the SEP and compare the activity between them by evaluation of the ERD/ERS in different frequency bands (Della Penna et al., 2004).

(III) Examine lateralization: Compare the activity ipsilateral and contralateral to the stimulated nerve (Enatsu et al., 2014; Hirata et al., 2002; Nikouline et al., 2000)

(IV) Examine the excitation and inhibition of the SMC by quantification of the mu and beta ERD and ERS, respectively (Della Penna et al., 2004; Hirata et al., 2002; Neuper & Pfurtscheller, 2001b; Nikouline et al., 2000; G. Pfurtscheller et al., 2002)

(V) Study the relation between SEP and ERS/ERD (G. Pfurtscheller et al., 2002)

In this thesis, I will present a new use of MNS in research consisting on the study of the relation between brain oscillations in the SMC and motor behaviour. I will also show its potential to be used as a treatment for TS.

2.4.3. MNS used in the presented studies

In all studies presented in this thesis using MNS, the right median nerve was stimulated using Digitimer DS7A HV Current Stimulator (*Digitimer Ltd, Hertfordshire, Uk*). Stimulation was delivered using a bar electrode consisting of two stainless steel disk electrodes with a diameter of 8mm separated by 30mm placed on the right wrist of the participants. The intensity of the stimulation used was the minimum intensity at which a thumb twitch was seen. A conductive gel (*Signagel*) was used to avoid burning and increase conductivity. Figure 2.6 shows an image of the bar electrode attached to the wrist of a participant.

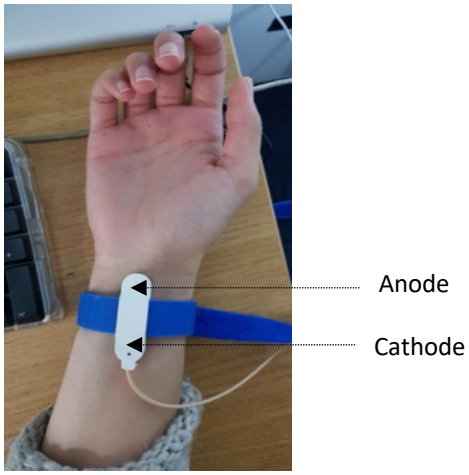


Figure 7.6. Image showing the placement of the electrodes

CHAPTER 3

Chapter 3 - Exploring the brain activity prior to tics and voluntary movements using EEG

CHAPTER 3A.1

Chapter 3a.1 - Exploring the brain activity prior to voluntary movements

Tics in TS are thought to be the result of the spontaneous firing in the striatum leading to an overexcited motor cortex. As discussed in Chapter 1, the execution of voluntary movements can suppress tics. In the first section of this chapter (i.e. Chapter 3a.1), I will explore the state of the motor cortex prior voluntary movement in HS and subjects with TS using EEG and present evidence of an altered motor cortex in subjects with TS prior and during voluntary movement.

3a.1.1. Introduction

ERP and ERSP prior and during voluntary movement

A voluntary movement is defined by a premotor potential (also called Bereitschaftspotential (BP) or readiness potential (RP)), a motor potential (MP) and a post-movement potential. The premotor potential is divided in two different components. First, there is the pre-movement potential, also called early BP, which is represented by a negative wave beginning about 1.5 or 2 seconds before the movement. Then, there is a more rapidly rise of negativity at about -500ms, called late BP or negative slope, mainly localized to the contralateral M1. The MP is represented by a positive peak and occurs with the movement onset and is localized to the contralateral M1 and SMC. Finally, the MP is followed by a positive post-movement potential. Previous studies have

shown that voluntary movements produced by subjects with TS are also preceded by BP (Karp, Porter, Toro, & Hallett, 1996; Obeso & Marsden, 1981; Van Der Salm, Tijssen, Koelman, & Van Rootselaar, 2012).

A voluntary movement has an effect in mu and beta oscillations in the motor cortex. A decrease in power (i.e. ERD) in both rhythms starts shortly before the movement and lasts for the whole movement execution. After movement is terminated, there is an increase in power (i.e. ERS) in both rhythms. ERD is thought to reflect cortical activation, while ERS is thought to reflect reduced cortical excitability or inhibition. This is supported by TMS studies, which have shown that there is an increase in cortex excitability just before a movement (80-100ms before movement) (Chen et al., 1998), during a movement (Chen et al., 1998); and decrease cortex excitability between 500-1000ms after the movement (Chen, Corwell, & Hallett, 1999).

Previous EEG/MEG studies have shown that voluntary movements produced by subjects with TS are also characterized by a mu and beta ERD and post-movement mu and beta ERS (Franzkowiak et al., 2010; Niccolai et al., 2016; Laura Zapparoli, Macerollo, Joyce, Martino, & Kilner, 2019). However, studies have reported reduced beta ERD in the contralateral SMC when performing externally driven movements compared to HS (Niccolai et al., 2016; Laura Zapparoli et al., 2019). No difference between HS and subjects with TS has been found in the alpha band when executing a voluntary movement (Franzkowiak et al., 2010; Laura Zapparoli et al., 2019). TMS studies have shown reduced increased cortical excitability in the motor cortex during movement preparation compared to HS (Draper, Jude, Jackson, & Jackson, 2015; Heise et al., 2010; S. R. Jackson et al., 2013); and MRI studies have shown a decreased BOLD signal in the motor cortex during execution of movement compared to HS (Roessner et al., 2012; Thomalla et al., 2014). A neuroimaging meta-analysis including 14 task-related fMRI studies with subjects with TS (Polyanska, Critchley, & Rae, 2017) reported a correlation between motor cortex activation and tic severity. It has been suggested that this increase in inhibition of motor cortical excitability is necessary in order to

generate the desired response and control the execution of tics (Draper et al., 2015).

Summary and aim

In summary, individuals with TS show a reduced motor cortical excitability prior to and during voluntary movement compared to HS. The aim of this study is to examine the changes in spectral power of mu and beta oscillations in the motor cortex preceding voluntary movement in HS and in subjects with TS. The reason to use a computer task instead of a simple finger movement task was to distract participants from ticcing in order to collect clean EEG data. The reason I did not compare the data between HS with subjects with TS was due to the TS subjects' heterogeneous sample (age, medication, comorbidities, etc.) and the fact that I did not recruit matched controls.

3a.1.2. Methods

Participants

HS: 20 participants (13 females) between 19 and 48 (mean age of 24.1) were recruited from the University of Nottingham. One participant was left-handed. All participants signed the consent form of the study, approved by the Ethics Committee in Nottingham University and they were given 10 pounds for their time. There were no requirements to take part in the study.

TS: 14 participants (6 females) between 12 and 51 (mean age of 22.5) were recruited for the study. Three participants were left-handed. All participants had a confirmed diagnosis of TS. Participants were recruited from our clinic database and *Tourette's Action*. All participants (or a parent/guardian) signed the consent form of the study, approved by the Ethics Committee in Nottingham University and they were given 10 pounds for their time. Participants with comorbidities were not excluded.

Two TS participants' data were excluded due to excessive noise caused by the frequent and intense tics. After rejections, the 12 participants now included 5 females between 12 and 33 (mean age of 20.3). Three participants were left-handed. Of the twelve subjects, 7 were taking medication, as follows: one was taking Sulpiride (antipsychotic), two were taking Clonidine (antihypertensive), one was taking Citalopram (antidepressant), one was taking Paroxetine (antidepressant), one was taking both Clonidine and Risipiradone (antipsychotic), and one was taking Aripriprazole (antipsychotic), Circadin (melatonin receptor agonist).

WASI scores varied from 82 to 130 with an average of 104.83. The total YGTSS score (100/100) varied from 23 to 92 with an average of 58.33 and the YGTSS score without taking into account their self-impairment measure (50/100) varied from 23 to 42 with an average of 32.08. Their PUTS (32/32) score varied from 7 to 30 with an average of 21.17.

Results of the scores can be found in Table 3a.1.1.

Table 3a.1.1. Participant characteristics

TS ID	Age	Gender	Handedness	Medication	Comorbidity	WASI	PUTS (max32)	Impairment*	Motor*	Vocal*	Global*
TS184	19.37	M	RH	Sulpiride 400mg	None	82	30	50	21	21	92
TS212	15.44	F	LH	None	Ehler Danlos syndrome	110	29	49	19	13	81
TS182	18.25	F	LH	Clonidine 4x25micrograms/day	None	85	18	20	21	9	50
TS213	21.39	F	RH	Citalopram 20mg/day	None	109	22	45	22	16	83
TS030	22.34	M	RH	None	None	103	7	17	15	8	40
TS204	33.19	M	RH	Clonidine 125 micrograms/day	None	130	15	5	18	15	38
TS229	32.97	M	RH	Paroxetine	TS	114	25	15	15	8	38
TS230	20.16	F	RH	None	TS	108	18	0	15	8	75
TS141	12.68	M	RH	None	TS	129	51	20	13	14	62
TS207	15.28	M	RH	None	TS	97	12	20	13	10	23
TS232	13.73	M	RH	Rispiradone 3mg/day, Clonodine 35micrograms/day	TS, ADHD	88	13	2	20	19	41
TS224	19.54	F	LH	Aripriprazole (25mg), 2mg circadin	Autism, Meares-Irlens syndrome, Phonological processing difficulty, dyspraxia, hypermobility, IBS	103	14	23	21	21	77
Average	20.36	-	-	-	-	104.83	21.17	22.17	17.75	13.50	58.33

WASI: Wechsler's Abbreviated Scale of Intelligence. PUTS: Premonitory Urge for Tics Scale (max score is 32). ADHD: Attention deficit hyperactivity disorder.
**Score taken from YGTSS: YGTSS Global Tic Severity Scale.*

Design

Participants performed a Go/NoGo task on the computer using the index finger of their dominant hand. Trials consisted of a fixation cross in the centre of the screen that disappeared after 1 second, and this was followed by the appearance of the stimulus (either a square or a circle) appearing in the centre of the screen. The task consisted in pressing the space button when a yellow circle or a blue square appeared in the centre of the screen (i.e. Go condition) and withholding a response when a blue circle or a yellow square appeared in the centre of the screen (i.e. NoGo condition). 70% of the trials were Go trials while 30% of the trials were NoGo trials. The diameter of both stimuli was 5cm long. When no response was made, the next trial appeared after 1 second. When a response was made, the next trial began.

Before the start of the study, participants performed a short practice of 20 trials. If at least 90% of those trials were performed correctly, the subject proceeded to start the actual study; if less than 90% of those trials were performed correctly, the subject continued to perform another practice of 20 trials until at least 90% of those trials were performed correctly.

EEG set-up and data collection

EEG set-up and data collection were performed as explained in the General Methods.

Devices

The visual stimuli were presented on a computer screen (Samsung S23A700D, screen resolution: 1920 x 1080). Participants made responses on an Apple Pro Keyboard M7803 connected to a Mac Pro computer running High Sierra (v 10.13.6) via USB cable. The task was controlled by PsychoPy2 (v1.90.3) script coded using python2 on the Mac Pro.

Experimental procedure

Participants were seated approximately 70cm away from the screen. There were 400 trials and a short break was offered every 100 trials. After every break, a reminder of the instructions of the task appeared on the screen. The task did not take longer than 15 minutes. TS participants were told not to suppress their tics.

Pre-processing of the EEG data

Pre-processing of the EEG data was performed as explained in the General Methods. Time-windows of -1 to 1 seconds, time-locked to the button press were extracted in both studies.

HS: No more than one channel per subject was deleted, and no more than 4 channels per subject was interpolated. In total, only one channel was deleted from one participant. TS: No more than one channel per subject was deleted, and no more than 5 channels per subject were interpolated. In total, only one channel was deleted from one participant. All those epochs in which the participant was ticcing were discarded.

EEG data analyses

Data analyses of the EEG data was performed to obtain the average ERP and average ERSP of the contralateral motor cortex to the hand used to press space (i.e. C3 on right-handed participants and C4 on left-handed participants).

I was only interested in trials were there had been a volitional movement and therefore, all NoGo trials and Go trials were no button was pressed were

discarded from the analyses and focused on the Go trials were the button was pressed.

Statistical analyses were performed using a one-sample t-test with a test variable of 0 in mu (8-13Hz) and beta (13-30Hz) at each time point, correcting for the error of multiple comparisons using False Discovery Rate (FDR; Benjamini & Hochberg, 1995). This was done to test the null hypothesis that the sample data are from a population with mean equal to zero at the 5% significance level.

3a.1.3. Results

HS: A minimum of 189 Go trials per subject and an average of 255.7 was obtained after cleaning the data. The button presses in the Go trials were performed at an average of 503ms, after the stimulus onset. TS: A minimum of 45 Go trials per subject and an average of 172.5 was obtained after cleaning the data. The button presses in the Go trials were performed at an average of 502ms, after the stimulus onset.

Event related perturbation (ERP) in HS

The ERP showed the BP and MP that characterises the voluntary movement (see Figure 3a.1.1). The BP starts at 550ms before the MP. The MP starts with the button press and ends 340ms after that. This is followed by a post-movement rebound.

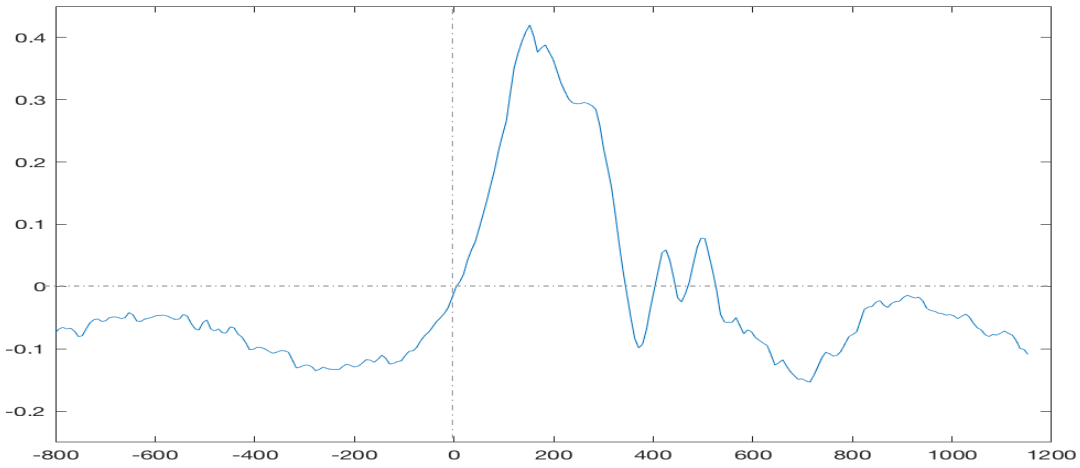


Figure 8a.1.1. ERP shown in the contralateral M1. Time 0 indicates the button press.

Event related perturbation (ERP) in TS

The ERP showed the BP and MP that characterises the voluntary movement (see Figure 3a.1.2). The BP starts at 535ms before the MP. The MP starts with the button press and ends 347ms after that. This is followed by a post-movement rebound.

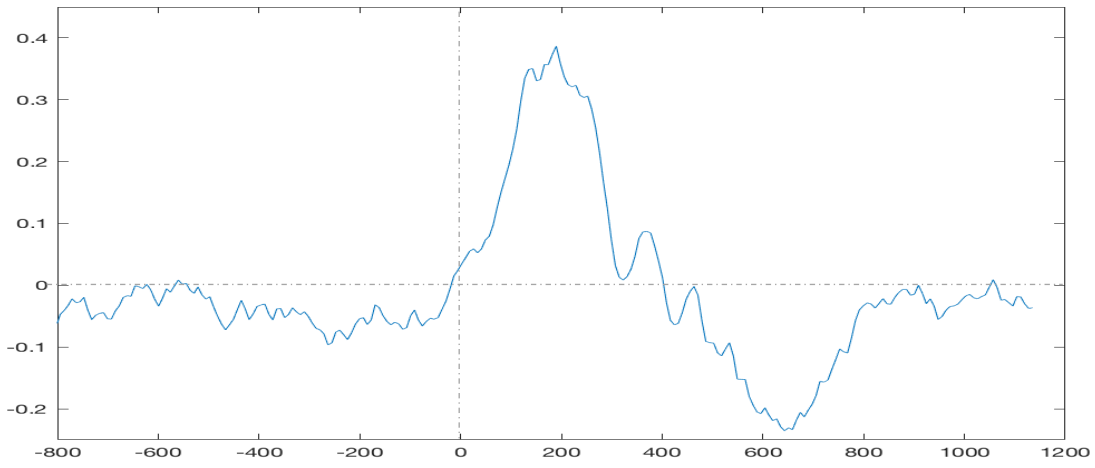


Figure 9a.1.2. ERP shown in the contralateral M1. Time 0 indicates the button press.

Mu rhythm in HS

The mu rhythm in the contralateral M1 showed a significant reduction in power (i.e. ERD) prior to the button press and during the execution of the finger movement (see Figure 3a.1.3). The significant ERD at 12Hz starts 20ms before the movement and ends 370ms after the button press.

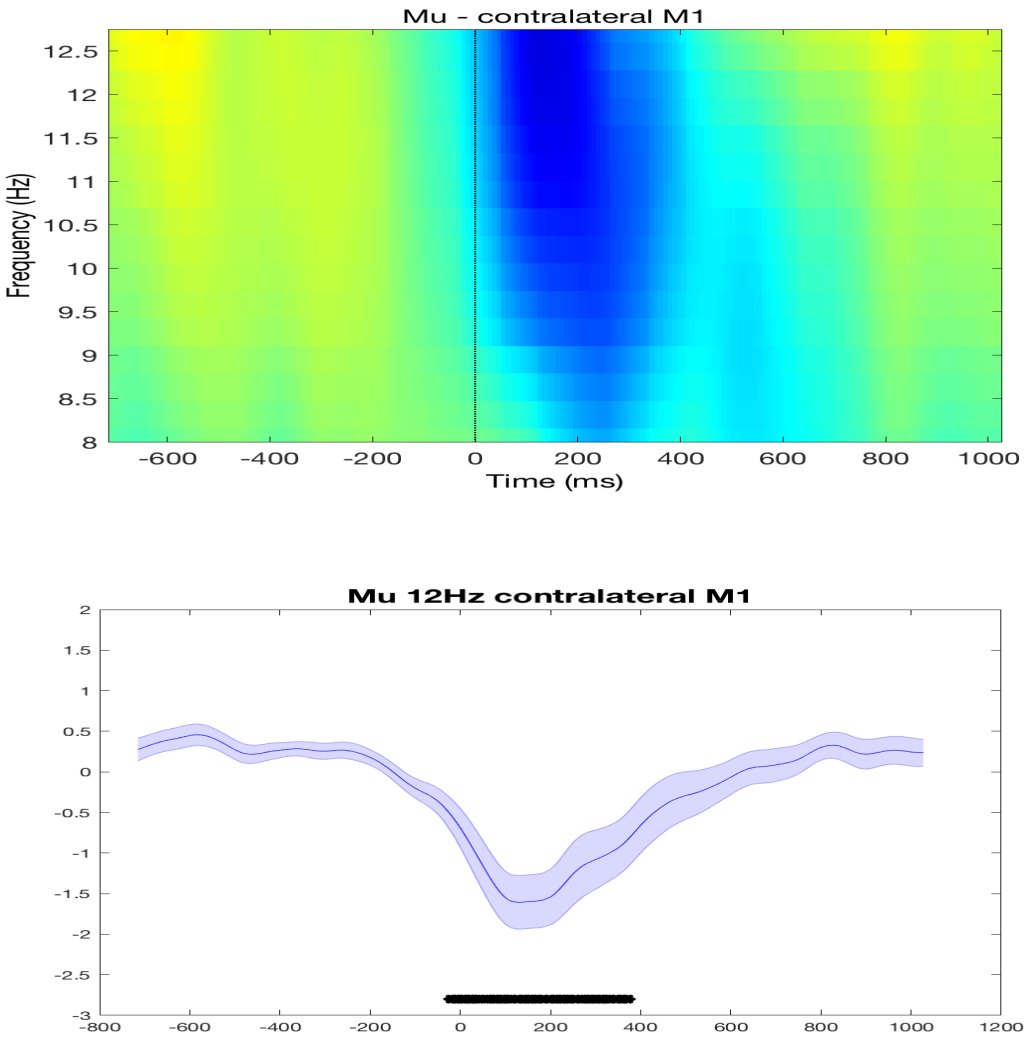


Figure 10a.1.3. A: Time-frequency analyses from the contralateral M1 in the mu band; B: Values of power at 12Hz recorded from the contralateral M1 in time. Time 0 indicates the button press. The black horizontal line indicates the time points were the values of power are significant.

Mu rhythm in TS

The mu rhythm in the contralateral M1 showed no significant reduction in power (i.e. ERD) prior to the button press and during the execution of the finger movement (see Figure 3a.1.4).

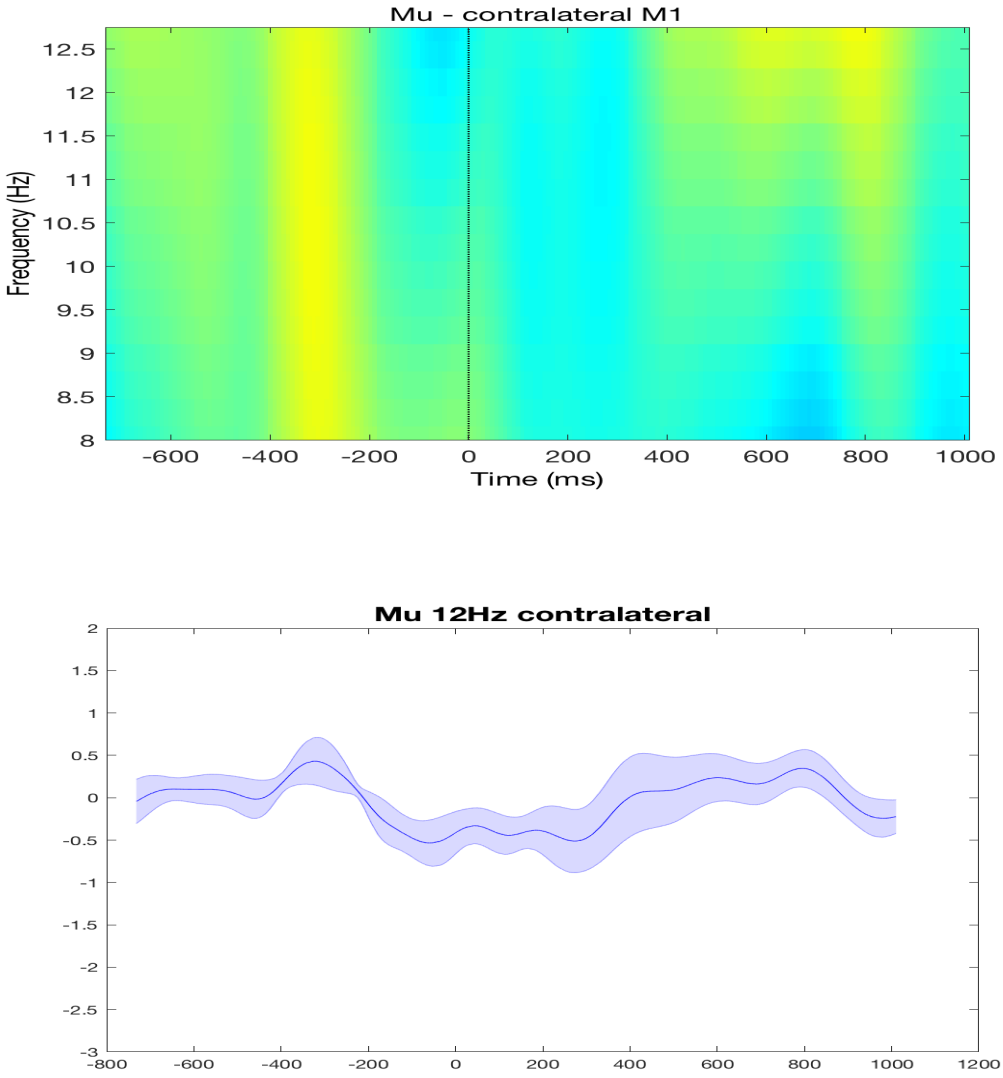


Figure 11a.1.4. A: Time-frequency analyses from the contralateral M1 in the mu band; B: Values of power at 12Hz recorded from the contralateral M1 in time. Time 0 indicates the start of the movement.

Beta rhythm in HS

The beta rhythm in the contralateral M1 showed a significant reduction in power (i.e. ERD) before the button press and during the execution of the finger movement (see Figure 3a.1.5). The significant ERD at 19Hz starts 191ms before the movement and ends 277ms after the button press.

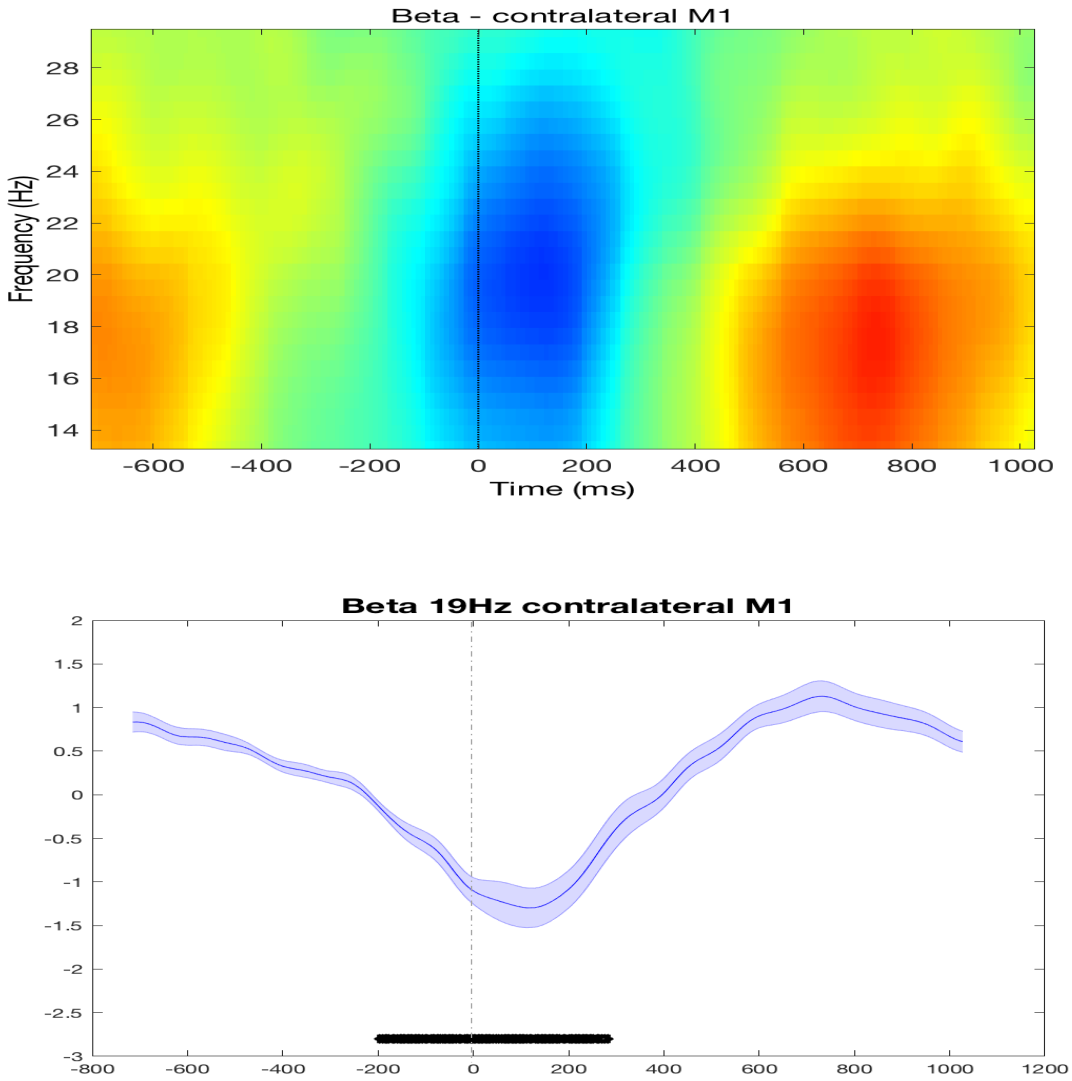


Figure 12a.1.5. A: Time-frequency analyses from the contralateral M1 in the beta band; B: Values of power at 19Hz recorded from the contralateral M1 in time. Time 0 indicates the start of the movement. The black horizontal line indicates the time points were the values of power are significant.

Beta rhythm in TS

The beta rhythm in the contralateral M1 showed a significant reduction in power (i.e. ERD) before the button press and during the execution of the finger movement (see Figure 3a.1.6). The significant ERD at 19Hz starts 113ms before the movement and ends 66ms after the start of the movement.

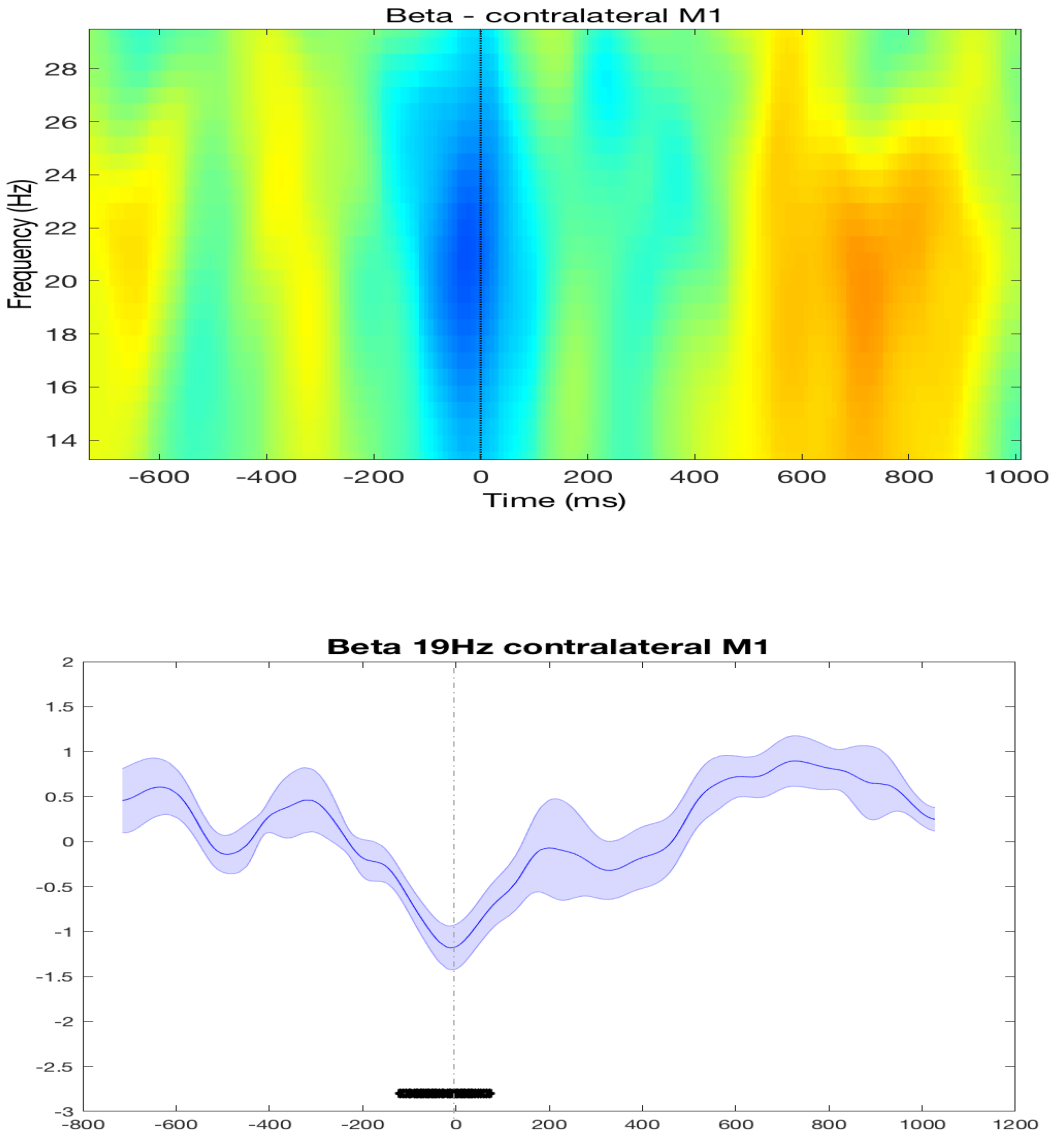


Figure 13a.1.6. A: Time-frequency analyses from the contralateral M1 in the beta band; B: Values of power at 19Hz recorded from the contralateral M1 in time. Time 0 indicates the start of the movement. The black horizontal line indicates the time points were the values of power are significant.

Power correlation with YGTSS *scores*

Mu power (12Hz) during the 15ms prior to the button press was significantly correlated (r=0.581, p=0.047) with the global YGTSS score (motor tic score + vocal tic score + impairment), indicating that the higher the mu power the higher the severity of the disease (see Figure 3a.1.7A). Beta power (19Hz) during the 15ms prior to the button press was not significantly correlated (r=0.099, p=0.759) with the global YGTSS score (motor tic score + vocal tic score + impairment) (see Figure 3a.1.7B).

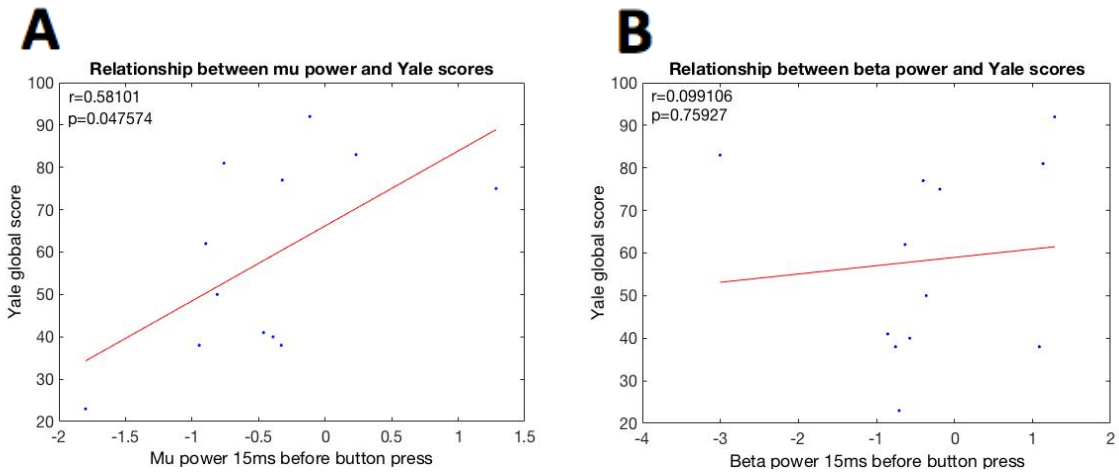


Figure 1415a.1.7. A: Relationship between the Mu power prior to button press and the global YGTSS scores. B: Relationship between the Beta power prior to button press and the global YGTSS scores.

Mu power (12Hz) at the time of the button press was significantly correlated (r=0.632, p=0.027) with the global YGTSS score (motor tic score + vocal tic score + impairment), indicating that the higher the mu power the higher the severity of the disease (see Figure 3a.1.8A). Beta power (19Hz) at the time of the button press was not significantly correlated (r=0.170, p=0.597) with the global YGTSS score (motor tic score + vocal tic score + impairment) (see Figure 3a.1.8B).

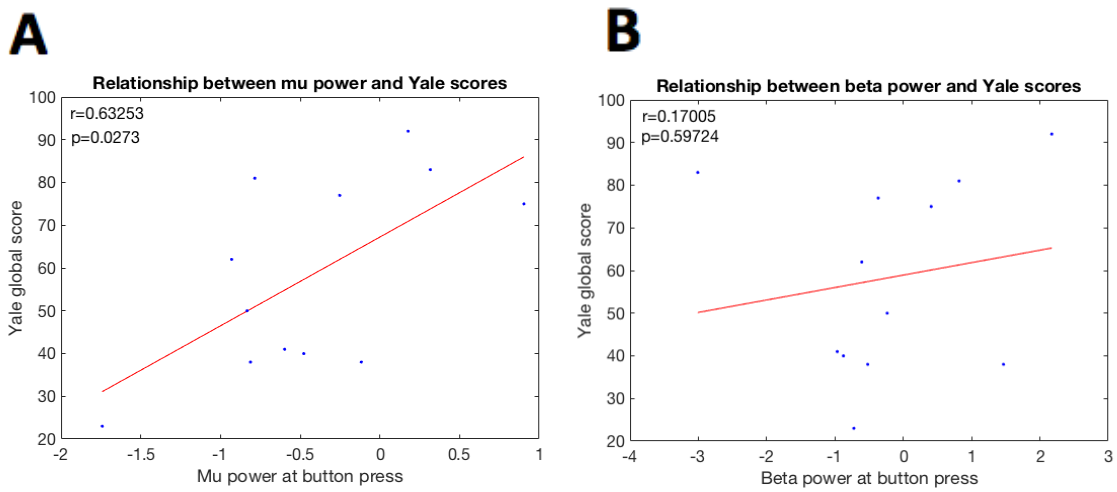


Figure 16a.1.8. A: Relationship between the Mu power at the time of button press and the global YGTSS scores. B: Relationship between the Beta power at the time of button press and the global YGTSS scores.

3a.1.4. Discussion

The aim of this study was to explore the cortical excitability preceding voluntary movements in HS and subjects with TS. Overall, results show that, as expected, voluntary movement is characterized by a BP, an MP and a post-movement potential in the contralateral motor cortex in both HS and subjects with TS; and that voluntary movement in HS is characterized by a decrease in power in both the mu and beta rhythms in the contralateral motor cortex starting shortly before the initiation of the movement. However, results also show that voluntary movement in subjects with TS is characterized by a decrease in power in the beta rhythm in the contralateral motor cortex starting shortly before the initiation of the movement, but no decrease in power in the mu rhythm prior or during voluntary movement.

On one side, these findings support previous results that have shown the presence of beta ERD in subjects with TS during an externally driven voluntary movement (Niccolai et al., 2016; Laura Zapparoli et al., 2019), although, as power differences between HS and subjects with TS were not analysed, I cannot confirm whether this decrease in beta power in subjects with TS was

significantly lower than beta power in HS, as reported by these two referenced papers.

On the other side, these findings contradict previous results that have shown the presence of alpha ERD in subjects with TS during an externally driven voluntary movement (Laura Zapparoli et al., 2019). However, this study recruited unmedicated participants, suggesting, as the authors comment in their limitations' section, that their sample might be part of a milder TS spectrum, which could be the cause of the dissimilarities in the results. In fact, our results showed that less severe participants present a stronger decrease in mu power than more severe participants during voluntary movement. The fact that TS subjects did not show an increase in excitability in the mu band preceding and during voluntary movement suggests that mu rhythm is involved in the previously reported reduced excitability preceding and during voluntary movement, and therefore indicates that this abnormal excitability is not specific to the beta rhythm, as stated by Zapparoli et al. (2019).

Mu power at the time prior to the button press as well as at the time of button press was positively correlated with tic severity: subjects showing a strongest increase in mu power were characterized by higher tic severity while subjects showing a strongest decrease in mu power were characterized by lower tic severity. Beta power was not significantly correlated with tic severity at the time prior to the button press or at the time of button press, supporting previous results (Niccolai et al., 2016). The fact that mu power at the time prior to button press and at the time of button press is correlated with tic severity suggests that subjects with severe tics require higher inhibition in the mu band in order to perform the desired voluntary movement than subjects with less severe tics.

Limitations

We did not apply baseline-correction to the data. Baseline correction involves subtracting the data found in an activity-free baseline time period within our selected time-window from the data of the whole time-window. By doing

this, we are shifting the overall data so that the baseline period is placed at 0 decibels. The period between stimuli appearance was short, therefore the activity preceding a voluntary movement within our selected time-window is likely contaminated with beta rebound, making it difficult to select an activity-free baseline time-period.

Because we did not apply a baseline-correction to our data, it is likely that the baseline is not placed at 0 decibels. This leads to uncertainty in estimating the onset or offset latencies of ERSPs. Because of this, these results should be interpreted with caution and considered as preliminary. I would suggest that in future experiments, the period between stimuli appearance is increased.

One might argue that the difference in the results between HS and subjects with TS is due to the different number of participants collected in each study. For this reason, in Appendix 1 of this thesis I have attached the analyses of the study with HS using the same number of subjects as the TS study (i.e. 12 subjects) as well as similar age (mean age of 21.8) and similar gender heterogeneity (6 females) showing similar results to the ones shown here.

3a.1.5. Conclusion and continuance

Our results show there is no increase in mu excitability in the contralateral motor cortex preceding as well as during voluntary movement in subjects with TS, suggesting an implication of mu rhythm in the characterized reduced cortical excitability preceding voluntary movement and during voluntary movement in subjects with TS, and therefore indicating that this reduced excitability is not specific to the beta rhythm, as previously reported. The present data also shows that the higher the mu power at the time prior to the movement execution as well as at the time of movement execution the more severity of the disease, suggesting that subjects with severe tics require higher inhibition in the mu band in order to perform the desired voluntary movement than subjects with less severe tics.

The second part of this chapter will explore cortical excitability preceding tics in subjects with TS.

CHAPTER 3A.2

Chapter 3a.2 - Brain activity prior to tics

Tics in TS are thought to be the result of the spontaneous firing in the striatum leading to an overexcited motor cortex. In this section, I will explore the state of the motor cortex prior to tics using EEG.

3a.2.1. Introduction

ERP prior to tics

EEG/MEG studies have examined the ERP of tics aiming to find a brain marker specific for tics that make them different from voluntary movements (e.g. no presence of BP), and an association between the ERP of tics and PU by looking at the presence or absence of BP in single subjects who report either the presence or the absence of PU.

Previous EEG studies examining the BP prior to the occurrence of tics have shown mixed results. Obeso & Marsden (1981) reported that BP was not observed preceding tics in 5 of the 6 participants; Karp, Porter, Toro, & Hallett (1996) reported that BP was observed preceding tics in 2 of the 5 participants; van der Salm et al. (2012) reported the presence of BP in 6 out of 14 patients; and Duggal et al. (2002) reported that BP was observed in all their 3 subjects, which they associated with experiencing PU.

ERSP prior to tics

Niccolai (2019) examined the changes in beta power in the SMC prior to tics using MEG, however, the analysed tics occurred while performing a Go/NoGo

task, and therefore, the data was likely contaminated by the effects of movement preparation and post-movement beta rebounds. To my knowledge, no other previous EEG/MEG studies have examined the changes in spectral power of brain oscillations prior to tics.

Summary and aim

There is mixed evidence of the presence of BP preceding tics that prevents from having a clear understanding of the voluntarity of tics and its association with PU. There is also a lack of knowledge about the state of excitability of the motor cortex just before tic execution by means of EEG.

I am interested in examining the changes in spectral power of mu and beta oscillations in the motor cortex prior to tics to identify a neural signature related to the initiation of tic execution. I will also examine the ERP of participants in order to provide further evidence of the presence or absence of BP before tic onset and its association with the experience of PU.

3a.2.2. Methods

Participants

Seventeen participants (5 female) between 12 and 26 (mean age of 18) with a confirmed diagnosis of Tourette Syndrome took part in the study. All participants had a confirmed diagnosis of TS. Participants were recruited from our clinic database and *Tourette's Action*. All participants (or a parent/guardian) signed the consent form of the study, approved by the Ethics Committee in Nottingham University and they were given 16 pounds for their time. Participants with comorbidities were not excluded.

One participant's data had to be rejected due to no intervals between tics. The 16 participants now included 4 females between 12 and 23 (mean age of 17.4).

Of the sixteen subjects, 5 were currently taking medication, as follows: one was taking Clonidine (antihypertensive), one was taking Aripiprazole (antipsychotic), one was taking Sulpiride (antipsychotic), and one was taking Clonidine (antihypertensive), Depakote (antiepileptic) and Quetiapine (antipsychotic). I only tested participants that presented few head jerks and very frequent facial and/or vocal tics within 30 minutes.

WASI scores varied from 82 to 139 with an average of 106.8. The total YGTSS score (100/100) varied from 23 to 92 with an average of 53.25 and the YGTSS score without taking into account their self-impairment measure (50/100) varied from 12 to 44 with an average of 29. Their PUTS (32/32) score varied from 4 to 30 with an average of 16.

Results of the scores can be found in Table 3a.2.1.

Table 3a.2.2. Participant characteristics

TS ID	Age	Gender	Medication	Comorbidity	WASI	PUTS	Impairment*	Motor*	Vocal*	Motor+Vocal*	Global*
TS030	21.1	M	None	None	103	5	10	13	11	24	34
TS061	14.8	M	Clonidine (75mg/day)	None	99	14	2	19	16	35	37
TS062	22.7	M	None	None	131	22	35	22	22	44	79
TS075	20.2	M	None	None		17	15	12	10	22	37
TS077	23.1	M	None	ADHD	91	8	10	17	18	35	45
TS139	14.4	M	None	None	100	5	25	12	0	12	37
TS141	12.7	M	None	None	129	10	35	13	14	27	62
TS171	18	F	None	None	101	9	8	9	6	15	23
TS176	12.5	M	None	None	107	4	17	12	13	25	42
TS178	18.2	F	Aripiprazole 10mg/day	None	110	25	30	17	20	37	67
TS182	17.4	F	None	None	85	24	15	17	9	26	41
TS183	12.5	M	None	None	139	11	35	18	18	36	71
TS184	19.4	M	Sulpiride 400mg	None	82	30	50	21	21	42	92
TS189	12.4	M	Depakote 750mg, Clonidine 0.15mg, Quetiapine 200mg	None	99	21	30	18	18	36	66
TS190	23.5	M	None	None	114	30	40	18	10	28	68
TS191	15.9	F	None	ADHD, OCD, GID	113	23	30	19	2	21	51
Average	17.4	-	-	-	106.86	16.12	24.18	16.06	13	29.06	53.25

*Greyed out area indicates missing score. WASI: Wechsler's Abbreviated Scale of Intelligence. PUTS: Premonitory Urge for Tics Scale (max score is 32). ADHD: Attention deficit hyperactivity disorder. OCD: Obsessive compulsive disorder. GID: Gender identity disorder. *Score taken from YGTSS: YGTSS Global Tic Severity Scale.*

EEG set-up and data collection

EEG set-up and data collection were performed as explained in the General Methods. Additionally, two EMG electrodes were placed over the *orbicularis oris*. These two electrodes were placed to record the exact onset of tics involving mouth and nose movements.

Experimental procedure

At the start of the video recording, participants were asked to look at the camera and blink five times. Considering the ease in identifying the initiation of blinks in both the EEG raw data and the video, this was done in order to later synchronize the EEG data with the video recording. Then, participants were asked to relax and not to suppress their tics. The testing took 28 minutes with three small breaks every 7 minutes.

Selection and marking of tic events

EEG data was synchronised with the video recorded during the testing. QuickTime 7 Pro player was used to play the videos from the testing. The synchronisation was not difficult to achieve due to the ease in finding the 5 consecutive blinks in the EEG data and the allowance of the QuickTime 7 Pro player to go from frame to frame every 40ms (maximum error range of 20ms).

Once the EEG was synchronised with the video recording, all those tics that happened in a period longer than 1 second from the last tic or from a voluntary movement were marked in the EEG data. This process was performed two times for each subject to ensure that all marked tics were actually tics, that started at the marked time and were not preceded by other tics or voluntary movements within one second. Specifically, only those tics that were preceded by at least a second of a resting state (no movement except for non-tic blinks) and that the start of the tic could be seen in the EEG data were marked.

Because of these requirements, many data were lost, as the majority of tics were followed by others in a chain separated by less than a second. Also, for young participants it was difficult for them to keep still and not perform voluntary movements for 28 minutes.

Pre-processing of the EEG data

Pre-processing of the EEG data was performed as explained in the General Methods. Time-windows of -1.2 to 0.5 seconds, time-locked to the tic onset were extracted. In total, only one channel was deleted from one participant.

Data Analysis

Data analyses of the EEG data was performed to obtain the ERP and ERSP of the electrodes over the SMC (i.e. average of C3, C1, C2, C4).

Statistical analyses were performed using a one-sample t-test with a test variable of 0 in mu (8-13Hz) and beta (13-30Hz) at each time point, correcting for the error of multiple comparisons using False Discovery Rate (FDR; Benjamini & Hochberg, 1995). This was done to test the null hypothesis that the sample data are from a population with mean equal to zero at the 5% significance level.

3a.2.3. Results

EEG data

Selected tics included mouth movements (with and without vocal sounds), eye movements and eye blinks, nose movements, head movements and whole-body movements.

The number of tic events after the epoch rejection had been performed varied from 17 to 82 between participants with an average of 42 (see Table 3a.2.2). Only in four participants the number of trials was under 30 and in two of these participants the number was under 20 (i.e. 17 and 18).

Table 3a.2.2 Participant ID with their corresponding number of trials

TS030	TS061	TS062	TS075	TS077	TS139	TS141	TS171	TS176	TS178	TS182	TS183	TS184	TS189	TS190	TS191	Mean	SD
63	17	48	38	70	41	84	18	33	24	30	72	47	22	32	36	42.18	20.41

Event related perturbation (ERP)

Figure 3a.2.1 shows the average ERP from the average electrodes on the SMC from all participants. The ERP is characterized by a slightly positive wave 770 to 320ms before the tic onset and an increase in negativity starting after that (i.e. 320ms before the tic onset). This negativity stops increasing with the onset of the tic (15ms after the onset) and becomes positive 140ms after tic onset.

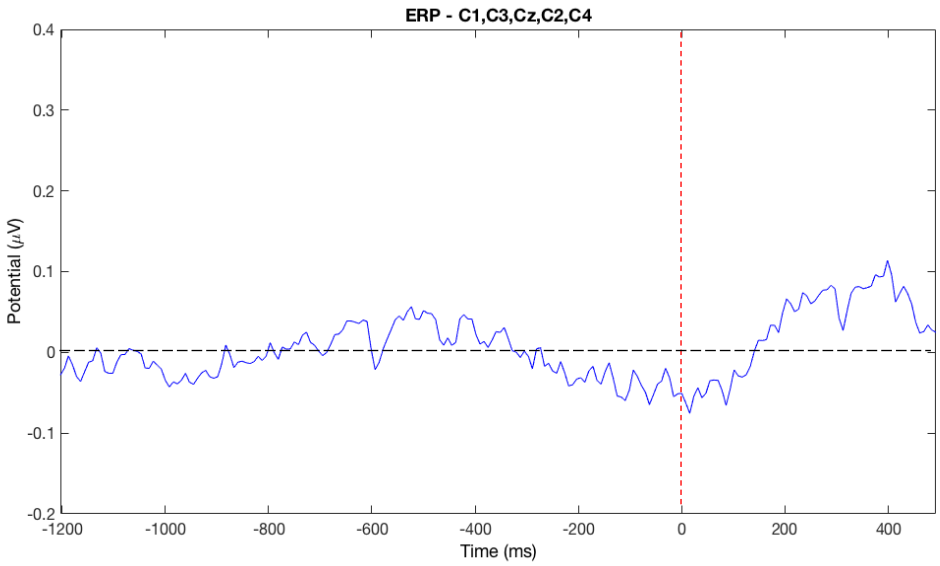


Figure 17a.2.1. Average ERP from the average electrodes on the SMC. Red line indicates the start of the tic.

Figure 3a.2.2 shows the average ERP for each of the subjects. Subjects TS030 and TS077 show a negative increase before the tic similar to a BP in voluntary movements.

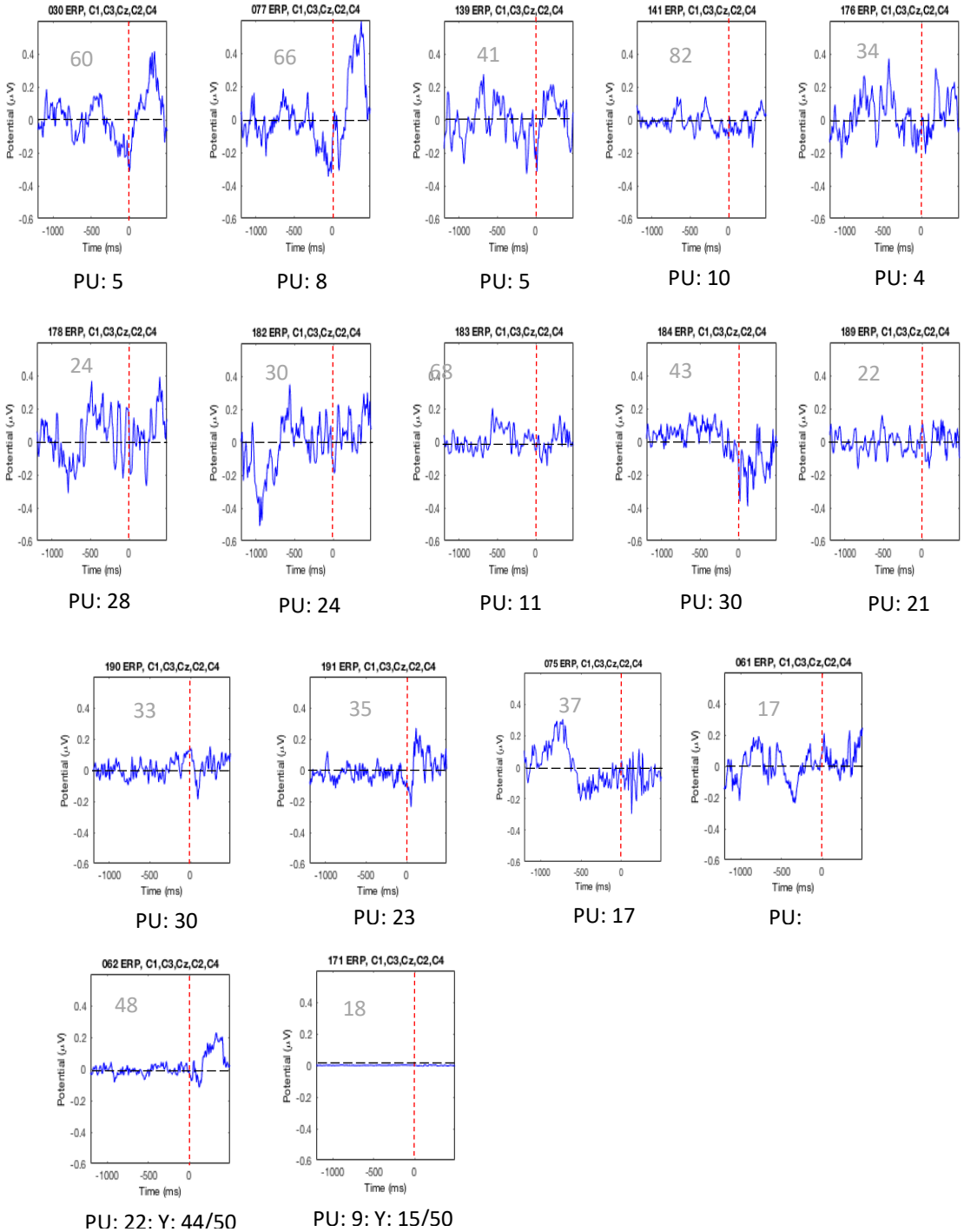


Figure 18a.2.2. Average ERP from the average electrodes on the SMC for each subject. Red line indicates the onset of the tic. Plots also show the number of trials (in grey) for each participant. PU: PUTS-R score.

Event related spectral perturbation (ERSP) of average trials

Time-frequency analyses were performed in alpha band (8-13Hz) and beta band (13-30Hz) for the average of the electrodes on the SMC.

Figure 3a.2.3 shows the time-frequency plots in alpha band for the average electrodes on the SMC. Results show a strong ERS starting shortly before tic onset that lasts for the rest of the epoch.

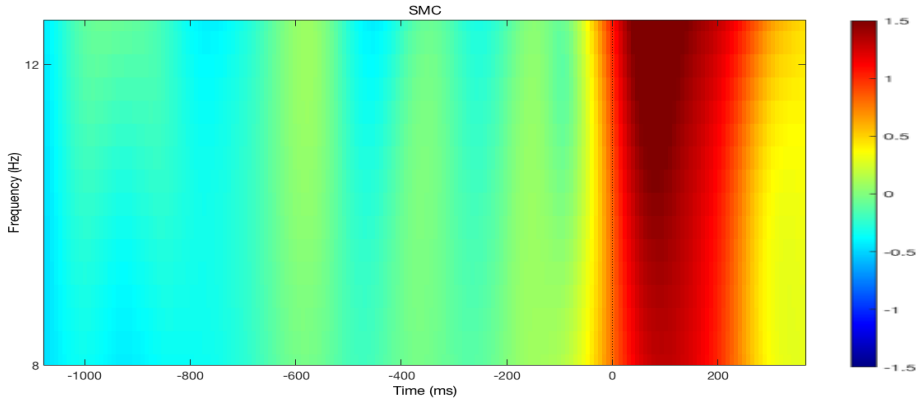


Figure 19a.2.3. Time-frequency plots in alpha band (8-13Hz) for the average electrodes on the SMC. 0 indicates the onset of the tic.

Figure 3a.2.4 shows the average ERSP values in the alpha band (8-13Hz) for the average electrodes on the SMC. Statistical analyses showed no difference between the ERSP and 0 ($p>0.05$, FDR corrected) in each timepoint prior to the onset of the tic. Statistical analyses showed no difference between the ERSP and 0 ($p>0.05$, FDR corrected) in each timepoint after the onset of the tic.

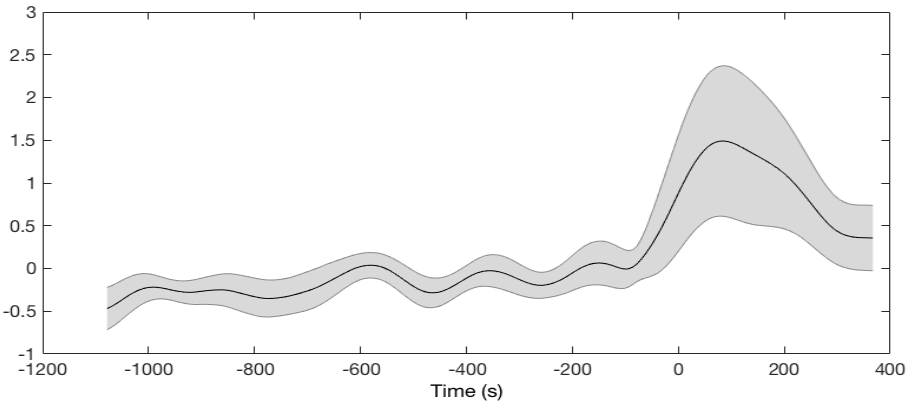


Figure 20a.2.4. ERSP values for the SMC in alpha band. 0 indicates the onset of the tic.

Figure 3a.2.5 shows the time-frequency plots in beta band (13-30Hz) for the average electrodes on the SMC. Results show a strong ERS starting shortly before tic onset that lasts for the rest of the epoch.

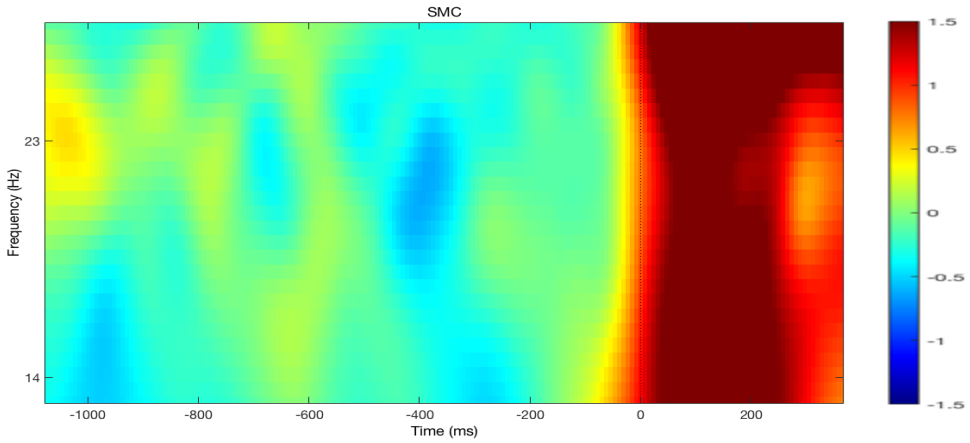


Figure 21a.2.5. Time-frequency plots in beta band (13-30Hz) for the average electrodes on the SMC. 0 indicates the onset of the tic.

Figure 3a.2.6 shows the average ERSP values in the beta band (13-30Hz) for the average electrodes on the SMC. Statistical analyses showed no difference between the ERSP and 0 ($p>0.05$, FDR corrected) in each timepoint prior to the onset of the tic. Statistical analyses showed no difference between the ERSP and 0 ($p>0.05$, FDR corrected) in each timepoint after the onset of the tic.

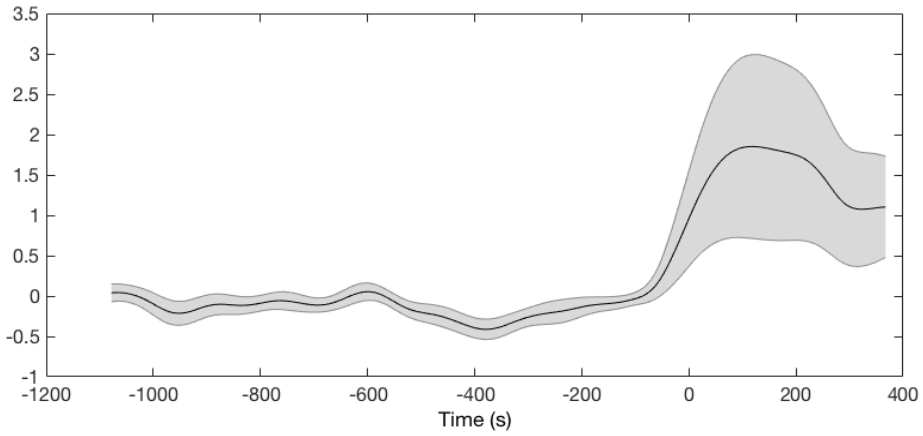


Figure 22a.2.6. ERSP values for the SMC in alpha band. 0 indicates the onset of the tic.

Event related perturbation (ERSP) of individual trials

Trials for each participant were rank-ordered to the strongest ERD peak before tic onset and this was then averaged between participants. This was performed in alpha band (8-13Hz) and beta band (13-30Hz) for the average of the electrodes on the SMC (see Figure 3a.2.7).

Results show a variability in time of the strongest peak of the ERD before tic onset that ranges from the start of the epoch (i.e. 1200ms before the onset of the tic) to the onset of the tic in both the alpha and beta frequency bands. Interestingly, the beta band shows more presence of ERD before tic onset and strongest ERS after tic onset.

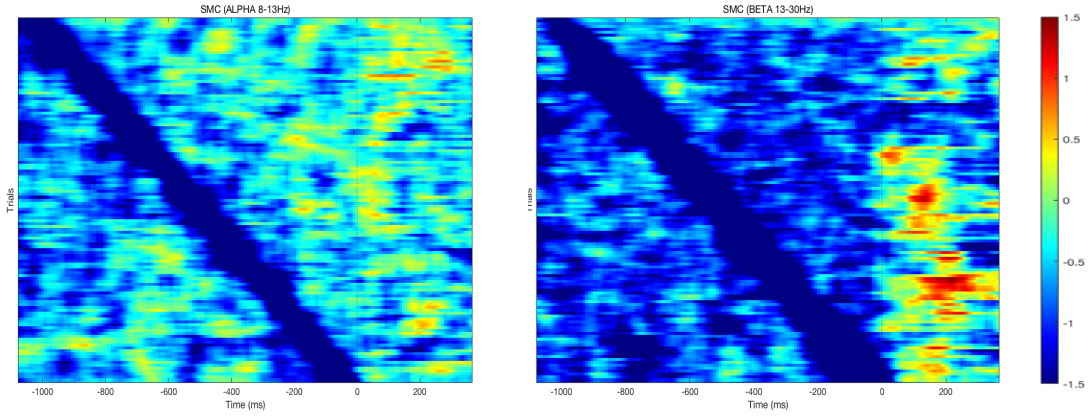


Figure 23a.2.7. Average of trials rank-ordered to the strongest ERD peak before tic onset in alpha band (left) and beta band (right) for the average electrodes on the SMC. 0 indicates the onset of the tic

3a.2.4. Discussion

ERP of averaged trials

BP preceding voluntary movements tends to be very consistent across HS (Gert Pfurtscheller, Zalaudek, & Neuper, 1998). It is seen in voluntary

movements of muscles in different parts of the body, like in movements of the sternocleidomastoid (Obeso & Marsden, 1981), deltoid (Karp et al., 1996) and trapezius (Karp et al., 1996; Obeso & Marsden, 1981), as well as in voluntary blinks (Berardelli, Rothwell, Day, & Marsden, 1985; Mota & Lins, 2017) and voluntary movements of the face like frowns and smiles (Recio, Shmuilovich, & Sommer, 2014). There were only two subjects from the 16 participants that showed a negative potential prior to tic onset similar to a BP preceding voluntary movements. The fact that PUTS score of these two participants was very low (5 and 8) suggest that the presence of BP is not associated with the presence of PU, contrary to Duggal (2002)'s conclusions.

On the contrary, BP is not seen in spontaneous movements in HS, like spontaneous blinks (Mota & Lins, 2017), or in blepharospasms (Berardelli et al., 1985), which are involuntary contractions of the eyelids. The fact that tics in fourteen of the sixteen subjects did not show a negative potential prior to tic onset similar to a BP suggest that tics are different from voluntary movement and similar to spontaneous, involuntary movements.

ERSP of averaged trials

Results show no sign of ERD just before the onset of a tic in the alpha or beta frequency bands, supporting the assumption that tics are different from voluntary movements.

The trials in the study included mainly simple tics. Simple tics have been found to be less detectable since a change in cortical signals are only seen in 39.2% of simple tics (Shute et al., 2016). Also, it is possible that since simple tics can be preceded by either a decrease or increase in the amplitude of brain oscillations in the motor cortex (Shute et al., 2016), the average of trials led to no change in power.

The ERS seen at the onset of the tic in the time-frequency analyses is not specific to a frequency band as it was seen in alpha, beta, and gamma, when

filtering from 8-63Hz (see Appendix 2), therefore it is likely that this ERS is due to movement artefact generated by the tic and not to brain activity.

ERSP of individual trials

Time-frequency of individual trials averaged between participants shows the variability in changes in spectral power that occurs before individual tics. Results show that there is in fact some level of ERD throughout the 1200ms before the tic execution in both the alpha and beta band, however, it does not seem to follow any consistent pattern. As expected, ERD in the beta band is found to be more consistent through participants as beta band is more strongly linked to motor function (i.e. tics), which were common to all participants, while mu band is more strongly linked to sensory phenomena (i.e. urges), which were not common to all participants.

Only a few tics show ERS after tic onset, which is consistent with the fact that the ERS is produced by the movement generated by the tic as we would not expect any movement artefact from a tiny mouth movement or a blink, but we would expect a big movement artefact from a head jerk or whole-body movement.

Limitations

We did not apply baseline-correction to the data. Baseline correction involves subtracting the data found in an activity-free baseline time period within our selected time-window from the data of the whole time-window. By doing this, we are shifting the overall data so that the baseline period is placed at 0 decibels. The period preceding a tic within our selected time-window is likely contaminated with pre-tic evoked activity, making it difficult to select an activity-free baseline time-period.

Because we did not apply a baseline-correction to our data, it is likely that the baseline is not placed at 0 decibels. This leads to uncertainty in estimating the onset or offset latencies of ERSPs. Because of this, these results should be interpreted with caution and considered as preliminary. I would suggest that in future experiments, participants with longer periods of time between their tics are recruited and this is used as activity-free baseline time-period for baseline-correction.

The camera was not able to capture some tics (e.g. abdominal tensing, leg and food tics) which might have happened before the time-locked tic and within the extracted epoch, which could lead to interpreting data as activity preceding the tic when in fact is activity produced by the execution of another tic.

Data had to be cleaned thoroughly in order to reduce the noise and increase the signal to noise ratio. Since the data was highly contaminated with tics, it is possible that during this process the signal may have been considerably reduced in the data.

The different properties from the averaged tics were not taken into consideration (e.g. where the tic was being executed, whether the tic included vocal sounds, whether the tic was preceded by an urge), leading to a very heterogeneous sample of tics, which could have led to the high variability of the results.

3a.2.5. Conclusion

I was not able to find a neural signature for tics. Trials showed the presence of desynchronization within 1.2 seconds prior to the tic execution; however, they did not follow a consistent pattern. This could be due to the high amount of variability between the trials, likely caused by the heterogeneity of the sample of tics.

The lack of BP and movement-related ERD immediately before tics likely reflects an absence in movement preparation, consistent with the assumption that tics are spontaneous, involuntary movements.

CHAPTER 3B

Chapter 3b - Exploring the brain activity prior to a tic using fMRI

Tics in TS are thought to be the result of the spontaneous firing in the striatum leading to an overexcited motor cortex. Premonitory urges (PU) are associated with activity in the somatosensory cortex and very often are referred by subjects as the cause of their tics. In the second part of this chapter (i.e. Chapter 3b), I will explore the activity in the brain prior to tics and during PU in TS using fMRI.

3b.1. Introduction

Brain activity prior to tics

Stern et. al. (2000) was the first study examining the resting state of TS patients with motor and vocal tics. They used event-related positron emission tomography (PET) techniques combined with video/audio recordings of tics. They found that tic occurrence was highly correlated with increased activity in a set of neocortical (supplementary motor area (SMA), medial and lateral premotor cortices, dorsolateral-rostral prefrontal cortex, primary motor cortex (M1), inferior parietal cortex, SMC, superior temporal gyrus, Broca's area), paralimbic (insula, anterior cingulate cortex (ACC)) and subcortical regions (claustrum, putamen, caudate nucleus).

After that, only two studies, to my knowledge, have examined the neural activity in TS patients prior to tic onset using imaging techniques (Bohlhalter et al.,

2006; Neuner et al., 2014). These studies examined the blood-oxygen-level dependent (BOLD) response at a resting state in a 1.5T scanner controlling tics with video/audio recordings before tic onset and at tic onset. They showed that 2 seconds before tic onset, there is activation in the SMA, parietal operculum (PO) (Bohlhalter et al., 2006; Neuner et al., 2014), primary sensory cortex (Neuner et al., 2014), thalamus, ACC and insula (Bohlhalter et al., 2006). Neuner et al. (2014) also reported activity in the ACC and insula, together with the putamen, amygdala, cerebellum, and occipital cortex 1 second before tic onset.

Brain activity during PU

PU is characterized by being an uncomfortable bodily sensation that precedes a tic and is experienced as a motivation for action (S. R. Jackson, 2011). This uncomfortable sensation is represented in the somatosensory cortex. When the action of the urge is suppressed, it is thought that the insula, which is the area that perceives bodily states, sensors the urge and recruits the ACC in order to suppress the action (S. R. Jackson, 2011; Alicja Lerner et al., 2009). The SMA is thought to be a mediator between the urge and the movement execution (Rajagopal et al., 2013). SMA together with the insula and cingulate cortex are thought to be the neurobiological signature for premonitory urges (PU) (Cavanna, Black, Hallett, & Voon (2017)). In fact the insula, ACC and SMA activate before tics but this activity is reduced at tic onset (Neuner et al., 2014), just like urges are increased before tics but they are reduced with the execution of the tic.

Summary and aims

In summary, previous studies seem to agree in the involvement of the insula, ACC, and SMA in PU and tic execution, as they activate before tics. However, none of the previous studies have directly investigated the specific activity associated with PU. In this study, I asked participants to suppress their tics

while pressing a button whenever they felt a strong urge to tic. The aim was to look at the brain activity at the strongest PU in order to identify a pattern of brain activity associated with the motivation for tic execution. Since participants were not very successful at suppressing their tics, I also examined the brain activity before tic onset in order to provide more evidence of brain areas activated before tics and report any differences between the activity at the strongest PU and the activity before tics. I also examined the brain activity at tic onset.

3b.2. Methods

Participants

20 participants (14 males) between 12 and 32 (mean age of 19) were recruited for the study. All participants had a confirmed diagnosis of TS. Participants were recruited from our clinic database and *Tourette's Action*. All participants (or a parent/guardian) signed the consent form of the study, approved by the Ethics Committee in Nottingham University and they were given 10 pounds for their time. All participants were screened with an MR volunteer safety screening questionnaire that was send by email, once they agreed to take part in the study, as well as prior to the taking part in the study on the day of the testing. Participants with comorbidities were not excluded.

Nine subjects were excluded for the following reasons: one participant had braces, one participant was too scared to go inside the scanner, during one participant's testing there was a failure in the scanner protocol, 3 participants showed too much movement after the pre-processing, and 3 subjects showed very little amount of events (PU<5). After rejections, the 11 participants now included 7 males between 14 and 32 (mean age of 20). Of these 11 participants, 6 were currently taking medication, as follows: three were taking clonidine (antihypertensive), one was taking elvanse (stimulant), one was taking co-codamol (analgesic), and one was taking mirtazapine (antidepressant).

WASI scores varied from 84 to 131 with an average of 102. The total YGTSS score (100/100) varied from 21 to 94 with an average of 43.45 and the YGTSS score without taking into account their self-impairment measure (50/100) varied from 20 to 44 with an average of 28. Their PUTS (32/32) score varied from 4 to 30 with an average of 19.

Results of the scores can be found in Table 3b.1.

Table 4b.1. Participant characteristics

TS ID	Age	Gender	Medication	Comorbidity	WASI	PUTS	Impairment*	Motor*	Vocal*	Motor+Vocal*	Global*
TS182	17.63288	F	None	None	85	20	10	14	11	25	35
TS062	22.70411	M	None	None	131	18	33	18	20	38	71
TS077	23.5589	M	None	TS, ADHD	91	7	25	13	7	20	45
TS058	14.4	M	Elvanse 50mg	ADHD	96	18	20	14	10	24	44
TS194	19.32603	F	Clonidine 50 micrograms	None	114	21	20	11	13	24	44
TS199	13.25753	M	Clonodine 75 micrograms	None	85	4	2	13	7	20	22
TS061	15.00274	M	None	None	99	29	0	18	16	34	34
TS204	32.94521	M	Clonidine 50 micrograms	None	130	11	5	17	10	27	32
TS205	17.45753	M	None	None	96	24	0	21	0	21	21
TS198	25.13151	F	Co-codamol 30mg/500mg (6x)	Depression	84	28	50	22	22	44	94
TS209	19.65753	F	Mirtazapine 30mg	Depression, anxiety, BPD	111	30	25	19	12	31	56
Average	20.09	-	-	-	102	19.09	17.27	16.36	11.63	28	45.27

*WASI: Wechsler's Abbreviated Scale of Intelligence. PUTS: Premonitory Urge for Tics Scale (max score is 32). ADHD: Attention deficit hyperactivity disorder. BPD: Borderline Personality Disorder. *Score taken from YGTSS: YGTSS Global Tic Severity Scale.*

Data collection

The MRI protocol consisted of a 3D T1-weighted anatomical scan and fMRI acquisitions, using a 3T Ingenia scanner (Philips Medical Systems, Best, The Netherlands) and a 32-channel head coil. The anatomical scan used a slab-selective axial 3D magnetization-prepared rapid gradient echo (MPRAGE) sequence, with TE= 8.5ms, TI= 960ms, TR-per-line of 8.3ms, TR per inversion pulse of 3000ms, matrix size 256x224x180, 1x1x1 mm raw voxel size, flip angle = 8°, parallel imaging factor of 1.7 in both phase encoding directions, and scan duration of 225 seconds.

Five fMRI acquisitions were acquired, each consisting of fat-supressed single-shot echo-planar imaging (EPI), with TR/TE = 1000/35ms, flip angle 65 degrees, matrix size 64x64, field of view 208x208 mm, 3.5x3.5mm pixels, 45 interleaved axial slices with slice thickness of 3 mm and 0.25 mm gap, in-plane parallel imaging factor of 2 and slice multiband factor of 3, with 300 dynamic volumes.

Experimental procedure

The aim of the experiment was to find BOLD activity patterns on the brain that happened at the moment of strongest feeling of urge to tic as well as one and two seconds before tic execution.

There was a total of 5 fMRI runs. In run 1, participants were told to press a button every 20 seconds, this run was used to map non-tic motor response; in runs 2 to 5, participants were told to press a button whenever they felt a strong urge to tic. These runs were used to capture brain activity associated with the urge to tic. The activity from the first run was later subtracted from the subsequent fMRI acquisitions.

Prior to the study, participants practiced the task outside the scanner, by lifting their index finger whenever they felt a strong urge to tic. This practice helped

participants understand what they were asked to do and also confirmed that they had understood the task. Participants were also asked to constantly suppress their tics while they were inside the scanner. This was done to avoid big head movements in the data.

To monitor the onset of the participants' tics, an MRI compatible camera (MR-compatible camera "12M-i" with integrated LED light, MRC Systems GmbH, Heidelberg, Germany) was attached to the head coil and placed in front of the participant's face to record tics happening on the face. Moreover, in runs 2-5, the researcher was situated beside the participant in the scanner room and pressed a button (using a second trigger box) every time she saw a tic happening in the body of the participant.

Overall, timings from 3 different events were collected: button presses (run1), premonitory urges (PU) indicated by the participant using the participant's button box (run2-5), and tics (TIC) indicated by the researcher using the researcher's button box (run2-5) and from the video camera (run1-5). Those PU button presses that happened at the same time as a tic (no more than half a second) were considered TIC events. In cases where the participant showed a chain of tics separated by less than 3 seconds, only the first tic of the chain was selected as TIC. Based on the TIC onsets, TIC-1 and TIC-2 onsets were created. TIC-1 were timing onsets one second before the TIC event, and TIC-2 were timing onsets two seconds before the TIC event.

MRI image processing and analyses

Data was extracted from the scanner in a DICOM format and was later converted to NIFTI format using MRIcron software (www.mricro.com). All subsequent analyses were performed using FSL (v. 6.00).

First of all, both the structural image and the 4D images of all runs were visually inspected to ensure that they had the correct orientation, correct labels (Right, Left, Anterior, Posterior) and no presence of artefacts. Skull strip of the

structural image was done with Brain Extraction Tool (BET) (Smith, 2002) with a fractional intensity threshold of 0.4. With the use of a mask, it was ensured that the skull strip had been performed correctly (i.e. non-brain had been removed and brain had not been removed). After that, using FSL Motion Outliers, those time points with movements bigger than 0.9mm, determined by the Framewise displacement (FD), were selected to be removed from the main parameter estimates.

All subsequent analyses were performed using FEAT (fMRI Expert Analysis Tool) version 6.00, part of FSL (FMRIB's Software Library, www.fmrib.ox.ac.uk/fsl).

Single run processing and analyses

Registration of the subject's functional image to the subject's high-resolution structural image was performed using Boundary based registration (Greve and Fischl, 2009). As a within-subject registration, it uses 6 degrees of freedom. Without going into detail, BBR creates a white-matter segmentation of the structural image and maps it to the EPI image; then the EPI image is transformed using the intensity voxels at both sides of the structural white-matter boundaries and the EPI intensity voxels. Registration of the high-resolution structural to standard space was performed using FLIRT (Jenkinson 2001, 2002), and was then further refined using FNIRT nonlinear registration (Andersson 2007a, 2007b). MNI152_T1_2mm was used as the standard space. As a between-subject registration, it uses 12 degrees of freedom.

The following pre-statistics processing was applied; motion correction using MCFLIRT (Jenkinson, 2002); non-brain removal using BET (Smith, 2002); spatial smoothing using a Gaussian kernel of FWHM 5mm; grand-mean intensity normalisation of the entire 4D dataset by a single multiplicative factor; high-pass temporal filtering (Gaussian-weighted least-squares straight line fitting, with sigma=50.0s). Slice timing correction was not needed due to the short TR of 1 second.

Time-series statistical analysis was carried out using FILM with local autocorrelation correction (Woolrich, 2001). The time series model included 5 regressors coding for button press, PU, TIC, TIC-1, and TIC-2 in 3 different GLMs/design matrices: (1) button press, PU, TIC events; (2) button press, PU, TIC-1 events; and (3) button press, PU, TIC-2 events. This was done to avoid unreliable parameter estimates and improve specificity, due to the high collinearity of the TIC, TIC-1 and TIC-2 regressors. The duration of the trials was set to 0 seconds due to their short latency.

The standard motion parameters, including the three translation parameters and the three rotation parameters, plus derivatives were included in the model as nuisance regressors. Volumes (i.e. single TR's) identified to have excessive motion according to FD>0.9, were excluded from the model – a separate regressor was used for each volume with high motion. Temporal derivatives were also added to the model. Double-gamma was used to convolve the HRF of the regressors, and temporal high-pass filtering was applied to the design to remove low-frequency noise. Finally, each regressor was contrasted to baseline (button press > baseline, PU > baseline, TIC > baseline, TIC-1 > baseline, TIC-2 > baseline).

Single subject analyses

Runs were rejected when timepoints exceeded 2mm, determined by the absolute mean displacement. If run 1 was rejected, the whole subject was then rejected. A total of 1 run was rejected from 6 different subjects. Rejected runs were not included in this level of analyses.

The second-level analysis was carried out using a fixed effects model, by forcing the random effects variance to zero in FLAME (FMRIB's Local Analysis of Mixed Effects) (Beckmann, 2003; Woolrich, 2004; Woolrich, 2008). These analyses averaged contrasts estimates over runs within subjects and also

contrasted PU>bpress by subtracting button press (run 1) from the average PU of all runs (runs 2-5).

Group analyses

The group analysis was carried out using FLAME (FMRIB's Local Analysis of Mixed Effects) stage 1 (Beckmann, 2003; Woolrich, 2004; Woolrich 2008), comprising both fixed effects and random effects to average the COPEs PU > button press, TIC > baseline, TIC-1 > baseline, and TIC-2 > baseline of all subjects.

Z (Gaussianised T/F) statistic images were thresholded non-parametrically using clusters determined by Z>3.1 and a (corrected) cluster significance threshold of P=0.05 (Worsley, 2001). Both mean>0 and mean<0 were analysed. This resulted in the mean BOLD signal that was significantly higher (activation) and lower (deactivation) compared to baseline at the moment of the PU>button press, TIC, TIC-1 and TIC-2.

3b.3. Results

Number of events

The number of button-presses in the first run ranged from 9 to 22, with an average of 14. The total number of tics (face and body tics) ranged from 17 to 95 between subjects, with an average of 51. The total number of PU ranged from 9 to 62 between subjects, with an average of 30. This information can be found in Table 3b.2.

Table 5b.2. Collected data

Subjects	Button presses	PU	TICs	Rejected runs
TS058	14	29	40	run05
TS061	12	53	68	run04
TS062	15	36	41	run03
TS077	22	9	19	
TS182	15	19	54	run03
TS194	15	28	72	
TS198	14	12	17	run02
TS199	15	15	76	
TS204	9	62	95	
TS205	16	17	49	run05
TS209	15	50	31	
Average	14.72	30	51.09	-
Sum	162	330	562	-

The maximum number of volumes excluded from the model (as FD>0.9) in a participant was 40 out of 300.

The number of tics was significantly lower in the first run, compared to the other runs (F=4.358; p=0.006). The number of tics and the number of PU selected by the participants were similar in all the last four runs of the study. Detailed information can be found in Table 3b.3.

Table 6b.3. Number of facial tics and number of button presses associated with urges in each run

	TIC					PU			
	Run 1	**Run 2**	**Run 3**	**Run 4**	**Run 5**	**Run 2**	**Run 3**	**Run 4**	**Run 5**
TS058	5	4	2	7	6	15	8	5	7
TS061	13	25	28	24	20	20	13	18	19
TS062	0	2	8	9	19	9	13	5	9
TS077	1	0	1	1	1	3	1	3	2
TS182	3	14	19	13	15	6	6	7	6
TS194	5	15	12	8	18	13	6	5	4
TS198	7	5	4	5	9	-	4	3	5
TS199	7	25	12	13	12	8	3	2	2
TS204	3	4	2	6	5	14	21	17	10
TS205	10	15	15	11	-	2	6	9	-
TS209	0	4	6	7	13	9	11	14	16
Average	4.9	10.27	9.9	9.45	11.8	9.9	8.36	8	8

Greyed out area indicates the runs that were not included in the analyses due to either excessive movement (TS058, TS061, TS062, TS182) or technical problems (TS198 and TS205)

GLM analyses

PU did not result in a significant increase or decrease in the BOLD.

Two seconds before the tic resulted in a significant increase in the postcentral gyrus (BA3b R). Results are presented in Figure 3b.4 and Table 3b.7.

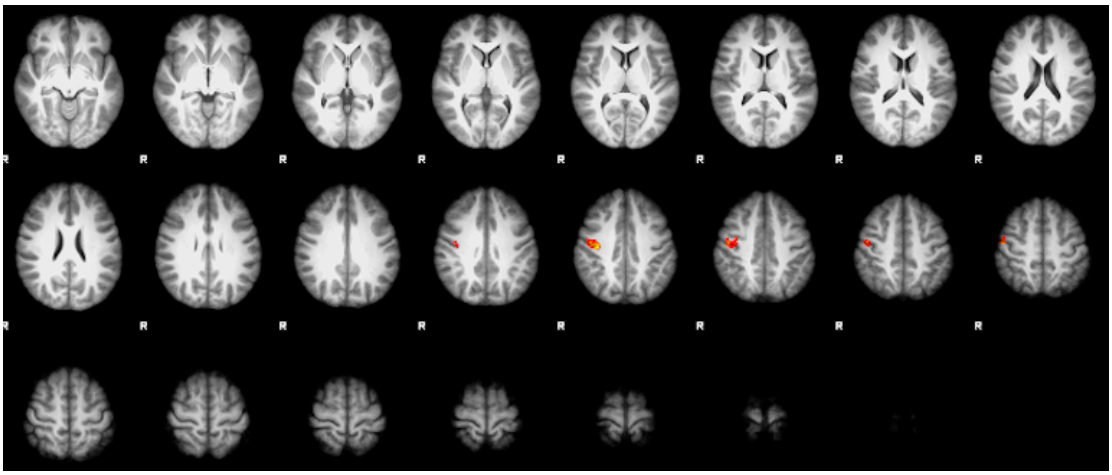


Figure 24b.1. Clusters exhibiting a significant BOLD increase corresponding to two seconds before tic onset (p<0.05 corrected for FDR) in whole-brain analyses

Table 7b.4. MNI coordinates and corresponding brain regions of statistically significant clusters of activation associated with button presses (p<0.05 corrected for FDR). Z-Max indicates the maximum z-score within the brain region.

	MNI coordinates							
	X	**Y**	**Z**	**Z-MAX**	**X**	**Y**	**Z**	**Z-MAX**
Brain region (Harvard-Oxford Cortical and Subcortical Structural atlases)	**Left hemisphere**				**Right hemisphere**			
Postcentral gyrus (Ba3b R)	-	-	-	-	38	-18	40	3.96

One second before the tic resulted in a significant increase in the right thalamus, and precentral gyrus (BA4p R). Results are presented in Figure 3b.3 and Table 3b.6.

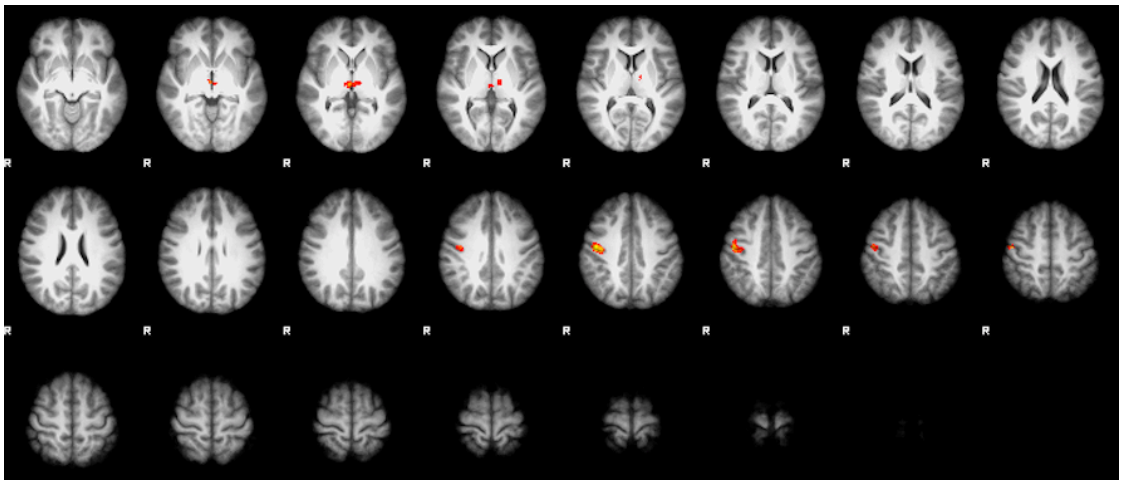


Figure 25b.2. Clusters exhibiting a significant BOLD increase corresponding to one second before tic onset (p<0.05 corrected for FDR) in whole-brain analyses

Table 8b.5. MNI coordinates and corresponding brain regions of statistically significant clusters of activation associated with button presses (p<0.05 corrected for FDR). Z-Max indicates the maximum z-score within the brain region

	MNI coordinates							
	X	**Y**	**Z**	**Z-MAX**	**X**	**Y**	**Z**	**Z-MAX**
Brain region (Harvard-Oxford Cortical and Subcortical Structural atlases)	**Left hemisphere**				**Right hemisphere**			
Right thalamus	**-**	-	-	-	4	-16	-2	3.79
Precentral gyrus (Ba4p R)	**-**	-	-	-	48	-12	42	3.98

Onset of the tic resulted in a significant increase in the precentral gyrus (BA 44L, BA4p L and BA4p R), cerebellum (Left V), frontal operculum cortex, and SMA. Results are presented in Figure 3b.2 and Table 3b.5.

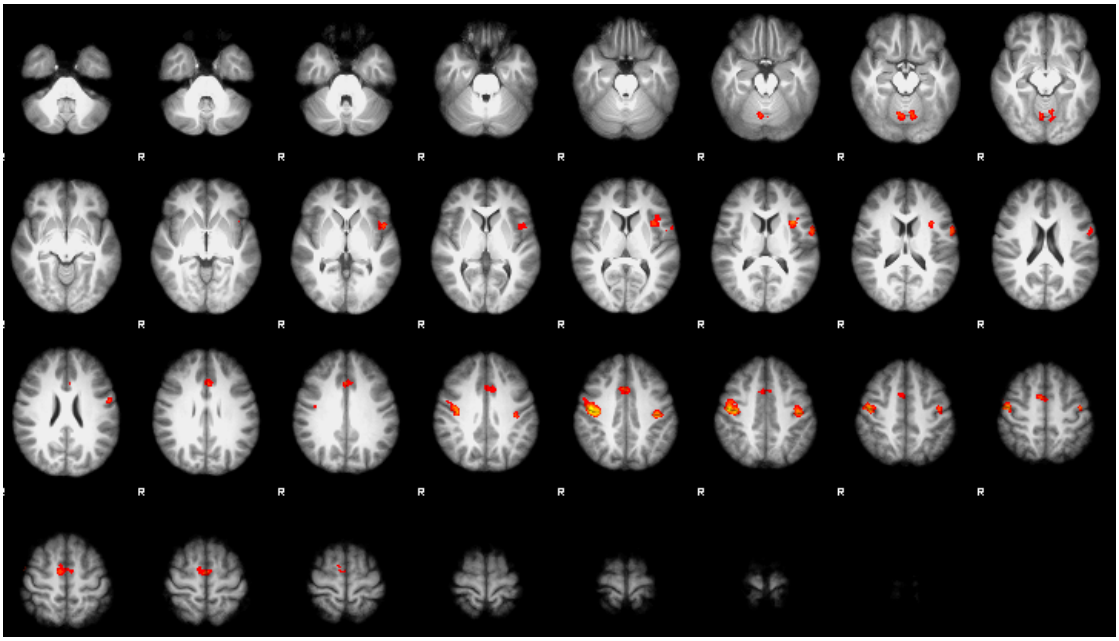


Figure 26b.3. Clusters exhibiting a significant BOLD increase corresponding to tic onset (p<0.05 corrected for FDR) in whole-brain analyses

Table 9b.6. MNI coordinates and corresponding brain regions of statistically significant clusters of activation associated with button presses (p<0.05 corrected for FDR). Z-Max indicates the maximum z-score within the brain region.

	MNI coordinates							
	X	**Y**	**Z**	**Z-MAX**	**X**	**Y**	**Z**	**Z-MAX**
Brain region (Harvard-Oxford Cortical and Subcortical Structural atlases)	**Left hemisphere**				**Right hemisphere**			
Precentral gyrus (BA44 L)	-60	4	14	3.72	-	-	-	-
Precentral gyrus (BA4p L) (BA4p R)	-40	-16	42	3.96	48	-12	42	4.57
Cerebellum	-6	-62	-14	3.65	-	-	-	-
Frontal operculum cortex	-34	12	12	3.96	-	-	-	-
Juxtapositional Lobule Cortex (formerly Supplementary Motor Cortex)	-	-	-	-	6	2	54	3.6

3b.4. Discussion

Brain areas activated before tics

Tics were preceded by an initial activity in the right postcentral gyrus (i.e. right primary somatosensory cortex) 2 seconds before tic onset, and this was followed by an involvement of the right thalamus and right precentral gyrus (i.e. primary motor cortex) 1 second before tic onset.

The involvement of the primary somatosensory cortex 2 seconds before tic onset supports Neuner's (2014) results and is likely related to the sensory phenomena, as previously suggested by Wang, Wells, & Wu's (2011).

The activity of the thalamus 1 second before tic onset supports the notion that there is a disinhibition of the thalamus that leads to tics. Also, the thalamus has been seen to activate when there is an urge to swallow or an urge to urinate (S. R. Jackson, 2011) and is part of the motivation-for-action system (S. R. Jackson, 2011).

The activity in the thalamus has also been seen during tic suppression (Peterson et al., 1998), as well as 2s before tic onset (Bohlhalter et al., 2006), were participants were aware that they should avoid head motion. It has also been reported at the onset of the tic (Neuner et al., 2014; Emily Stern et al., 2000), were participants were directly asked to not make any effort to suppress their tics. It is possible that the intention of the participants to suppress their tics leads to a short delay of the tic onset from the disinhibition of the thalamus.

The activity in the primary motor cortex may represent the intention of the participants to suppress the motivation for action (i.e. execution of the tic) evoked by the disinhibition of the thalamus. Increased activity in the primary motor cortex has also been seen during suppression of spontaneous blinking (Alicja Lerner et al., 2009).

Brain areas activated at the subjective strongest PU

There was no significant activity at the time of the strongest PU selected by the participants compared to baseline. Even though participants were asked to suppress their tics while inside the scanner, they were not successful. In fact, the total number of tics in the overall data was 562. Therefore, it is possible that the urge, which increases with suppression (Valerie C. Brandt et al., 2016), was not strong enough to elicit a significant increase in BOLD activity.

Differently to what was expected, there was no increase in activity in the right insula or the dorsal anterior cingulate cortex before tics or at the strongest PU. The activity in the insula correlates with the intensity of the itch (Roberts et al., 2019) as well as the intensity of the urge to tic (Tinaz, Malone, Hallett, & Horovitz, 2015), so it could be that the strongest the perception of the sensory states, the more the insula is activated. This could be a reason why the insula was not activated at the PU; the intensity of the urge was low as participants were not able to suppress their tics successfully and therefore, there was no increase in the intensity of the urge.

ACC is thought to contribute to the suppression of urges (Alicja Lerner et al., 2009). The fact that there was no significant activity in the ACC could be due to the fact that the participants were unable to successfully suppress their tics but also to the fact that the insula, which is supposed to recruit the ACC in order to suppress the urge, was not activated.

Brain areas activated during tics

The onset of these tics was associated with an increase in activity in the right SMA, cerebellum, left frontal operculum cortex, left precentral gyrus (BA44) and bilateral primary motor cortex (BA 4).

The SMA, primary motor cortex and cerebellum are known to be involved in movement generation and their activity has also been associated with tic onset

(Bohlhalter et al., 2006; A Lerner et al., n.d.; Neuner et al., 2014; Emily Stern et al., 2000; van der Salm et al., 2018). The activity in the frontal operculum cortex at tic onset has been reported before (Neuner et al., 2014; Emily Stern et al., 2000) and it was associated with coprolalia (Emily Stern et al., 2000). The frontal operculum is situated in the vicinity of Broca's area, and both areas cluster in the same Brodmann area (BA44); also, the activity in the left frontal operculum has been reported in fMRI studies involving language tasks and for these reasons it is said to be involved in language processing (Amunts & Zilles, 2012). Then, it is likely that the activity in the left frontal operculum and left precentral gyrus (BA 44L) are involved in the execution of vocal tics.

Limitations

Participants were characterized by having severe TS and most of them commented that their PU is always there and always strong, meaning that most of our subjects did not present an increase in urge intensity before tics and relief after the tic execution. It could be that the baseline was contaminated with PU and when subtracting it from the PU trials, it resulted in lack of activity.

3b.5. Conclusion

Main results show that tics are preceded by an initial activity in the right primary somatosensory cortex 2 seconds before tic onset, which can be related to sensory phenomena leading to the urge, and this is followed by an involvement of the right thalamus, which can be related to the urge to tic, and right primary motor cortex 1 second before tic onset, which can be related to the intention to suppress the urge to tic.

There was no significant activity at the time of the strongest PU selected by the participants compared to baseline, which can be explained by participants not feeling strong enough urges due to the short timing between their tics within the lasts runs. It could also be that the baseline was contaminated with PU.

The onset of the tics, including motor and vocal tics, led to activity in brain areas involved with movement generation and language processing.

CHAPTER 4

Chapter 4 - Exploring MNS as a tool to entrain the motor cortex

---

In the previous chapter, I have shown increased inhibition at mu rhythm in the motor cortex prior to and during voluntary movement in subjects with TS (Chapter 3a.1), as well as clear evidence of an inconsistent neural pattern in the motor cortex preceding tics involving a decrease in excitability within the second preceding the tic (Chapter 3a.2). Finally, in Chapter 3b, I have supported previous findings associating the primary somatosensory cortex with urges.

Overall, Chapter 3 has shown evidence of an aberrant SMC excitability prior to tics and voluntary movements. In the following chapter, I will show how it is possible to change the state of the SMC cortical excitability using a peripheral stimulation technique.

4.1. Introduction

Mu and beta rhythms are the strongest electrophysiological signals in the SMC and the desynchronization of these rhythms is related to an activated motor cortex while the synchronization of these rhythms is related to an inhibited motor cortex. In fact, the sensorimotor Mu desynchronization has been found to be positively correlated with corticospinal excitability (Thies, Zrenner, Ziemann, & Bergmann, 2018). The state of the cortex can be modulated by the application of an external rhythmic stimulation that leads to the phase alignment of the intrinsic neural oscillations with the external stimulation, generating entrainment.

Thut et. al (2011) delivered rhythmic trains using TMS while recording MEG and they used their results to define the entrainment signature. The entrainment signature was defined as an increase in power at the frequency of stimulation over the target area. They showed that in order for this entrainment signature to show, the stimulation initially has an effect on neural populations oscillating at many different frequencies and after a short number of pulses, the stimulation progressively starts recruiting only those neural populations oscillating at the frequency of stimulation, which phase align their oscillations to the pulses of stimulation, and that is when the entrainment signature is shown.

Entrainment increases the synchronization of brain oscillations resulting in an increase of cortical inhibition. Being able to modulate mu and beta oscillations in the motor cortex can be useful to treat motor conditions that show an altered motor cortex excitability, like Tourette syndrome (Heise et al., 2010). Non-invasive brain stimulation techniques (e.g. TMS or transcranial electrical stimulation (tES)) have been used to alter cortical excitability and modulate brain network function. However, these techniques require screening and are clinic based. Median nerve stimulation (MNS) is a non-invasive brain stimulation technique that does not require screening and can be applied outside the clinic and can be used to stimulate the SMC.

Summary and aim

In summary, evidence suggests that entrainment of brain oscillations in the SMC could have a beneficial effect in subjects with TS. MNS is a technique with characteristics that overcome the limitations from other currently used brain stimulation techniques. The aim of this study is to examine whether rhythmic MNS alters cortical excitability by showing the entrainment signature reported by Thut et al, (2011) on the contralateral motor cortex.

4.2 Mu entrainment study

4.2.1 Methods

Participants

Twenty healthy adult participants (7 males) between 18 and 38 (mean age of 24.2) took part in the study. One of the subjects was left-handed. There were no requirements to take part in the study. Participants were given 8 pounds for their time. Overall, 22 participants were recruited but one participant withdrew from the study due to feeling too much pain with the stimulation and another participant was not able to be tested due to not finding the threshold even at maximum intensity of stimulation. All participants signed the consent form of the study, approved by the Ethics Committee in Nottingham University.

EEG set-up and data collection

EEG set-up and data collection were performed as explained in the General Methods.

Devices

The stimulator was controlled by a bespoke MATLAB vR2017a script on a Mac Pro computer running High Sierra (v 10.13.6).

Experimental procedure

A number of 300 trains of 10 short pulses were delivered every 4 seconds to the right median nerve of the participants, while EEG was being recorded. The trains were delivered in two different conditions randomly. In the rhythmic condition, pulses were delivered every 83ms (i.e. 12Hz); in the arrhythmic

condition, pulses were delivered during the same time-window as the rhythmic condition but in random intervals of time. Figure 4.1 shows a scheme of the design of the study.

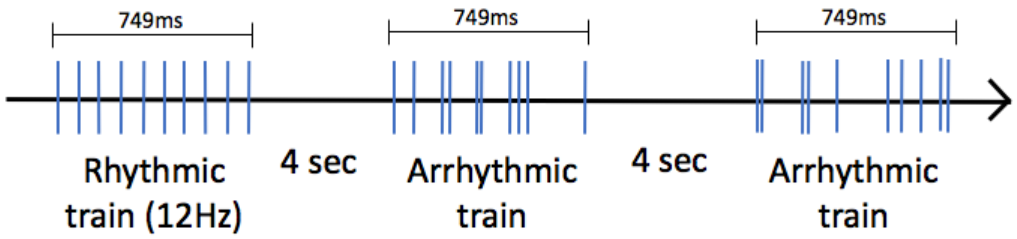


Figure 27.1. Scheme of the design of the study

The duration of a pulse was 200μs and the total length of the train was 749ms. Three short breaks were provided throughout the testing, one every 75 trains. Participants were asked to stay still, relax and keep their eyes open. Participants were monitored with a camera to make sure that the thumb twitch was always present. Intensity was adjusted if needed.

The starting intensity threshold was determined with a trial and error procedure with one pulse until a noticeable thumb twitch was seen.

Pre-processing of the EEG data

Pre-processing of the EEG data was performed as explained in the General Methods. Specifically, data was low-pass filtered at 30Hz and high-pass filtered at 8Hz, which included alpha and beta bands. Time-windows of -1 to 3 seconds, time-locked to the first pulse of the train were extracted.

In total, no more than 3 channels were deleted from all participants. The average number of epochs between participants after epoch rejection was 125 in the rhythmic condition and 123 in the arrhythmic condition.

Data Analysis

Data analyses of the EEG data was performed to obtain the ERSP and ITC; mu oscillations (8-13Hz), and scalp maps using whole scalp electrodes.

ERSP values and ITC were divided into window 1 (W1) and window 2 (W2) at frequency 11-13Hz. W1 corresponds to time 0 to 249, which includes pulses 1-3; and W2 corresponds to time 250 to 832, which includes pulses 4-10. W1 is plotted in black and W2 is plotted in red.

Statistical analyses were performed using a one-tailed paired-samples t-test correcting for the error of multiple comparisons using False Discovery Rate (FDR; Benjamini & Hochberg, 1995).

4.2.2 Results

All analyses were performed on electrode C1.

Event Related Spectral Perturbation (ERSP)

Time-frequency analyses showed an increase in power at the beginning of the train involving both mu and beta bands in both the rhythmic and arrhythmic conditions (see Figure 4.2A-4.2B).

After that, the rhythmic condition showed an increase in mu-power in a narrow band that peaked at the frequency of the stimulation (i.e. 12Hz) (see Figure 4.2B, W2). This increase in power centred at 12Hz was absent in the arrhythmic condition (see Figure 4.2A, W2). This difference becomes clearer when subtracting the arrhythmic condition from the rhythmic condition (see Figure 4.2C).

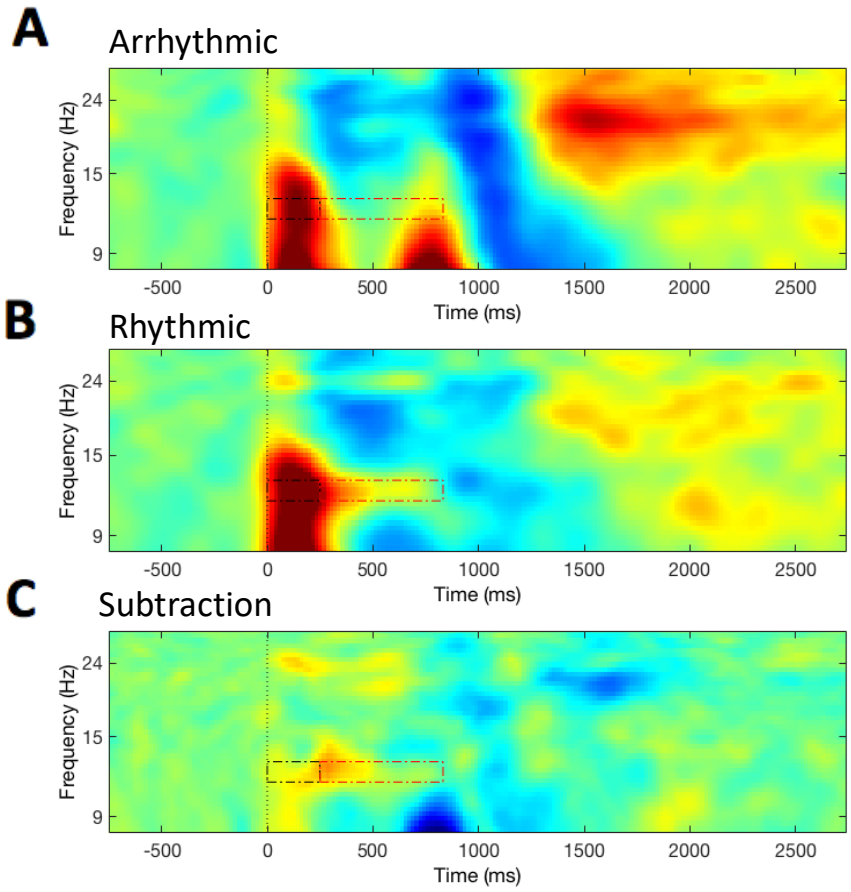


Figure 28.2. Time frequency analyses recorded from electrode C1 for A: the arrhythmic condition, B: the rhythmic condition, and C: the subtraction of the arrhythmic condition from the rhythmic condition. 0 indicates the time of the first pulse of the train. The black rectangle indicates W1 at 12Hz frequency and the red rectangle indicates W2 at 12Hz frequency.

Figure 4.3 shows the average of ERSP values recorded from electrode C1 at 12Hz for both conditions in time, with black lines indicating those time points where values were significantly different. Results showed that the increase in mu power at the frequency of stimulation specific of the rhythmic stimulation is significantly different from the arrhythmic condition. Specifically, significant difference ($p<0.05$) included timepoints ranging from 23ms to 78ms in W1 and from 289ms to 515ms in W2.

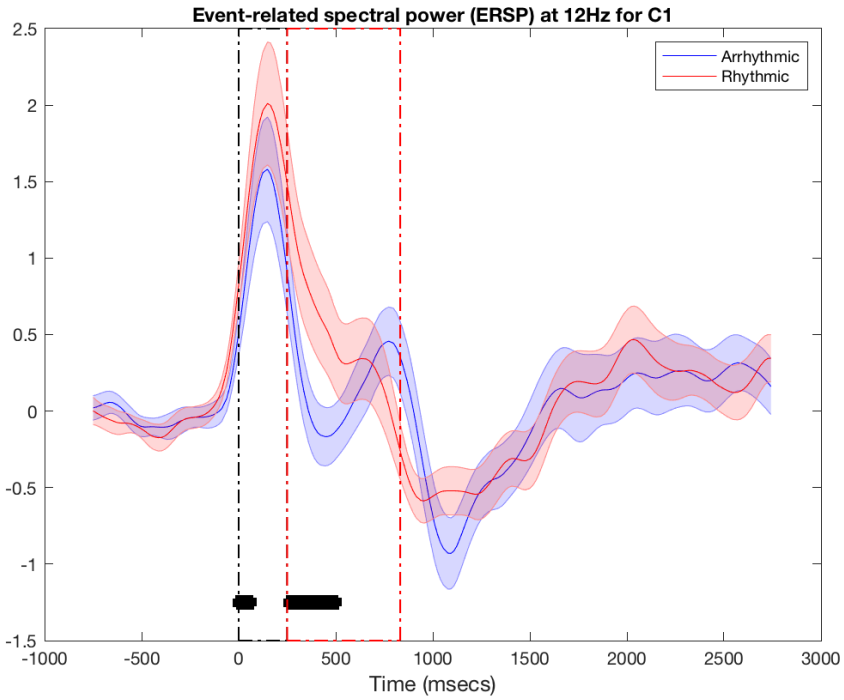


Figure 29.3. Time-frequency values recorded from electrode C1 at 12Hz for both the rhythmic (red) and arrhythmic (blue) conditions. 0 indicates the time of the first pulse of the train. Black lines indicate the time points where the time-frequency values between both conditions were significantly different. The black rectangle indicates W1 and the red rectangle indicates W2.

Inter-trial coherence (ITC)

ITC analyses showed an increase in phase locking at the beginning of the train involving both mu and beta bands in both rhythmic and arrhythmic conditions (see Figure 4.4A-4.4B).

After that, the rhythmic condition showed an increase in ITC in a narrow band that peaked at the frequency of the stimulation (i.e. 12Hz) (see Figure 4.4B, W2). This increase in phase locking centred at 12Hz was absent in the arrhythmic condition (see Figure 4.4A, W2). This difference becomes clearer when subtracting the arrhythmic condition from the rhythmic condition (see Figure 4.4C, W2).

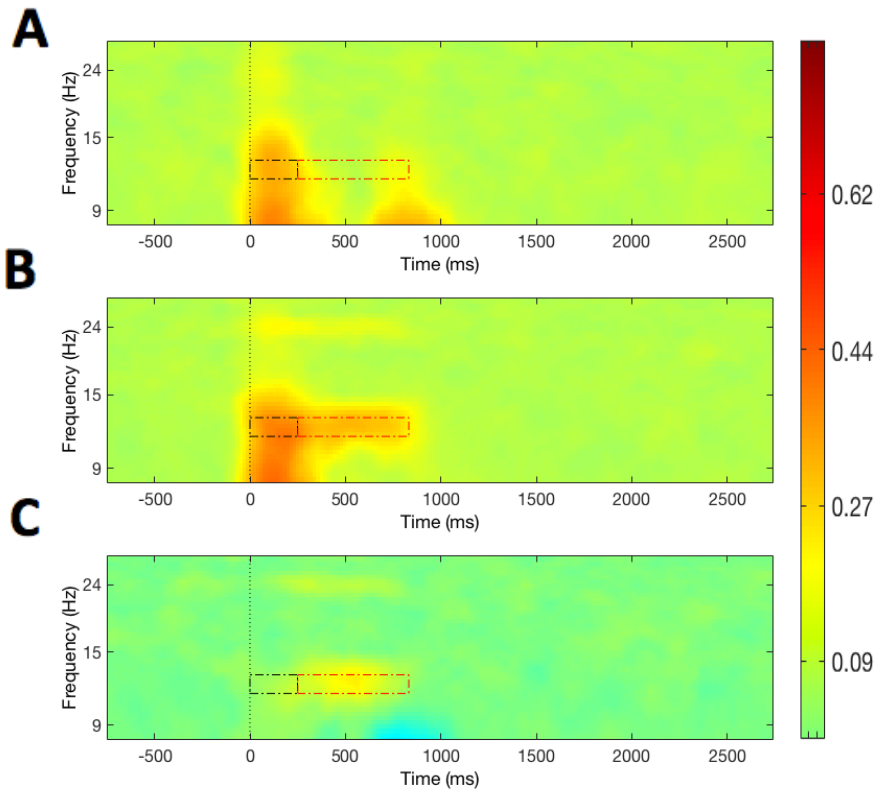


Figure 30.4. ITC analyses recorded from electrode C1 for A: the arrhythmic condition, B: the rhythmic condition, and C: the subtraction of the arrhythmic condition from the rhythmic condition. 0 indicates the time of the first pulse of the train. The black rectangle indicates W1 at 12Hz frequency and the red rectangle indicates W2 at 12Hz frequency.

Figure 4.5 shows the average of ITC values recorded from electrode C1 at 12Hz for both conditions in time, with black lines indicating those time points where values were significantly different. Results showed that the increase in ITC at the frequency of stimulation specific of the rhythmic stimulation is significantly different from the arrhythmic condition. Specifically, significant difference ($p<0.05$) included timepoints ranging from 179ms to 703ms.

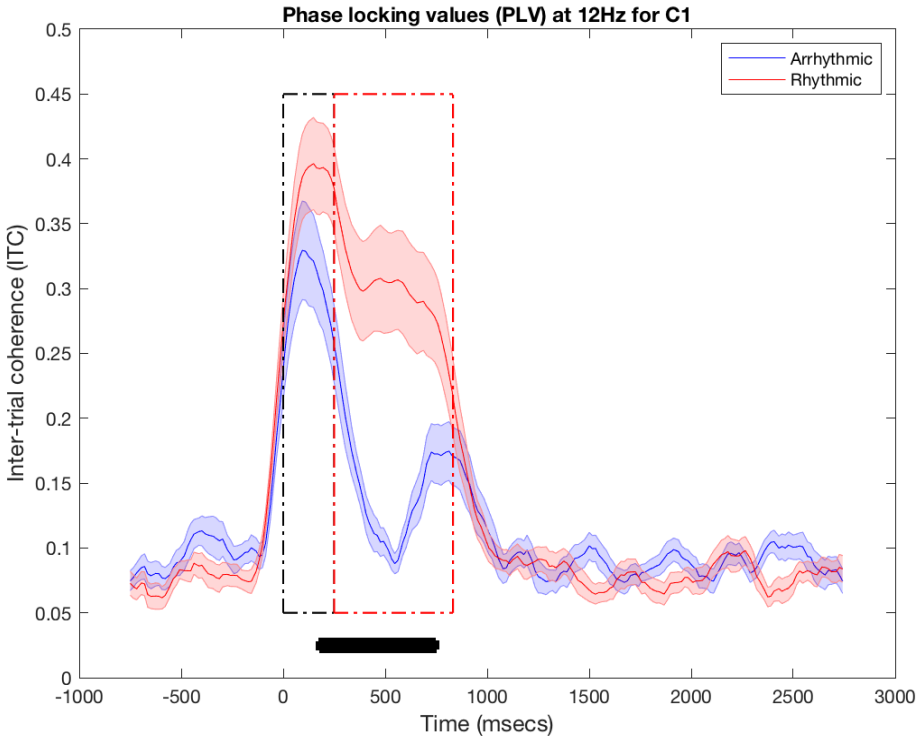


Figure 31.5. ITC values recorded from electrode C1 at 12Hz for both the rhythmic (red) and arrhythmic (blue) conditions. 0 indicates the time of the first pulse of the train. Black line indicates the time points where the ITC values between both conditions are significantly different. The black rectangle indicates W1 and the red rectangle indicates W2.

Inter-hemisphere effect

Figure 4.6A-4.6B shows the time-frequency analyses in the rhythmic condition from electrode C1 and C2. Electrode C1 is placed over the contralateral hemisphere to the stimulation while electrode C2 is placed over the ipsilateral hemisphere to the stimulation.

Results showed an increase in power at the beginning of the train involving both mu and beta bands in both C1 and C2 electrodes (see Figure 4.6-4.6B). After that, the contralateral hemisphere showed an increase in mu-power in a narrow band that peaked at the frequency of the stimulation (i.e. 12Hz) (see Figure

4.6A, W2). This increase in power centred at 12Hz was absent in the ipsilateral hemisphere (see Figure 4.6B, W2). This difference becomes clearer when subtracting C2 from C1 (see Figure 4.6C, W2).

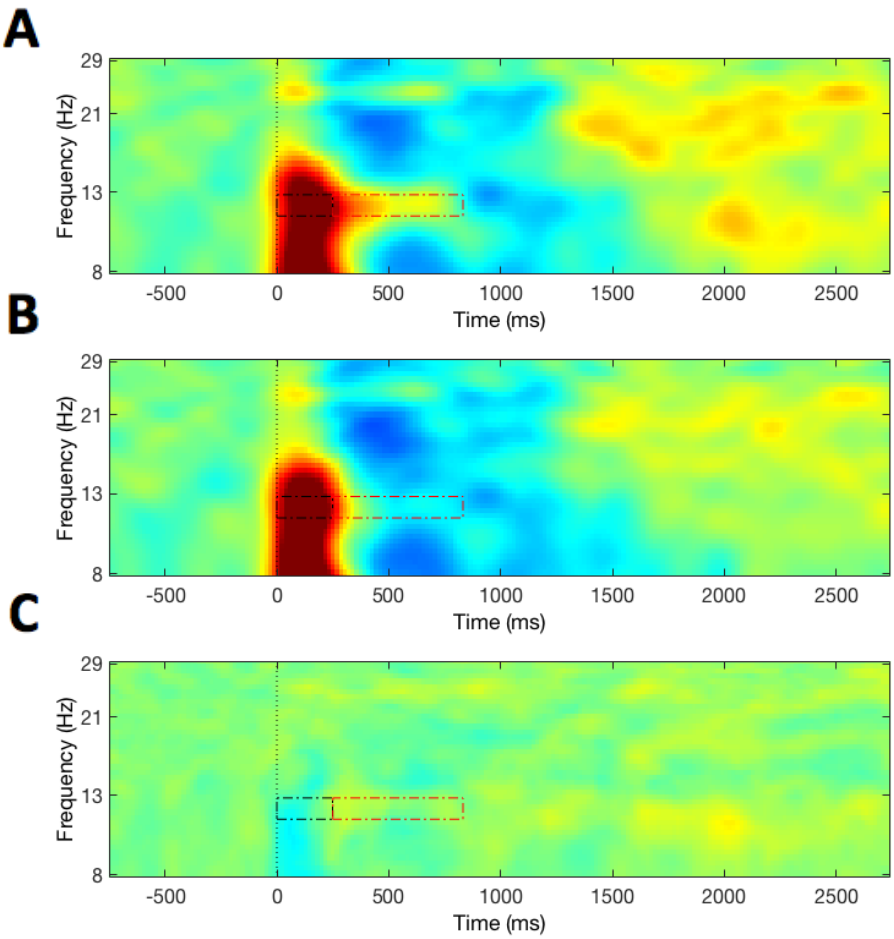


Figure 32.6. Time frequency analyses of the rhythmic condition for A: electrode C1, B: electrode C2, and C: the subtraction of the electrode C2 from the electrode C1. 0 indicates the time of the first pulse of the train. The black rectangle indicates W1 at 12Hz frequency and the red rectangle indicates W2 at 12Hz frequency

Figure 4.7 shows the average ERSP values recorded from electrode C1 and C2 at 12Hz in time, with black lines indicating those time points where values were significantly different. Results showed that the increase in beta power at the frequency of stimulation in C1 is significantly different from C2. Specifically, significant difference ($p<0.05$) included timepoints ranging from 304ms to 742ms. ERS power was also found to be significantly stronger in the arrhythmic condition compared to the rhythmic condition from 1953ms to 2148ms.

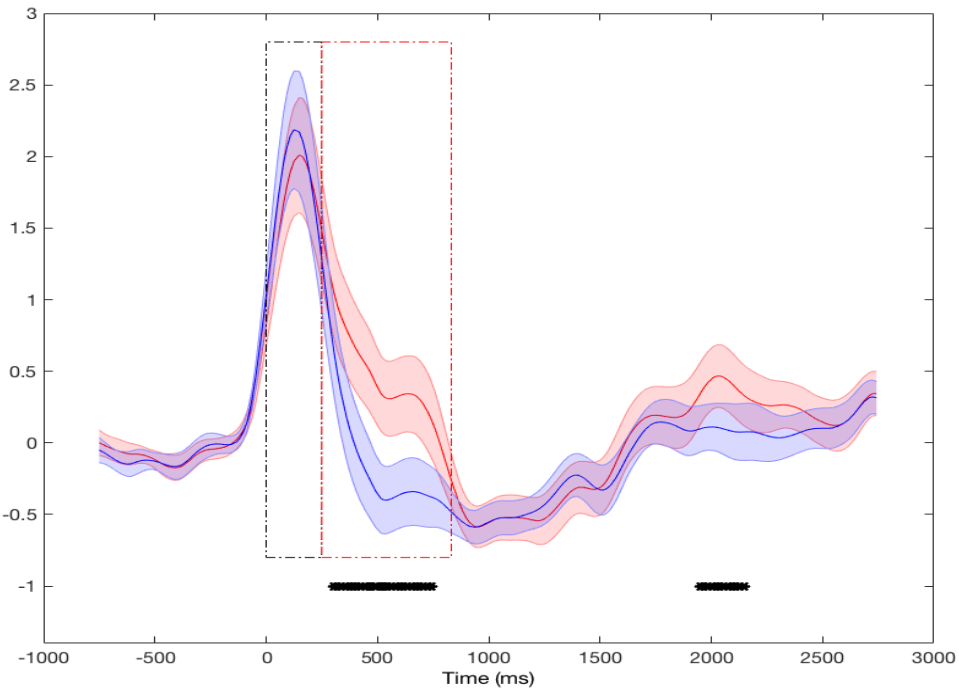


Figure 33.7. Time-frequency values recorded from electrode C1 (red) and electrode C2 (blue) at 12Hz for the rhythmic condition. 0 indicates the time of the first pulse of the train. Black lines indicate the time points where the time-frequency values between both electrodes are significantly different. The black rectangle indicates W1 and the red rectangle indicates W2.

Figure 4.8A-4.8B shows the ITC analyses in the rhythmic condition from electrode C1 and C2. Electrode C1 is placed over the contralateral hemisphere to the stimulation while electrode C2 is placed over the ipsilateral hemisphere to the stimulation.

Results showed an increase in phase locking at the beginning of the train involving both mu and beta bands in both C1 and C2 electrodes (see Figure 4.8A-4.8B). After that, the contralateral hemisphere showed an increase in ITC in a narrow band that peaked at the frequency of the stimulation (i.e. 12Hz) (see Figure 4.8A, W2). This increase in power centred at 12Hz was absent in the ipsilateral hemisphere (see Figure 4.8B, W2). This difference becomes clearer when subtracting C2 from C1 (see Figure 4.8C, W2).

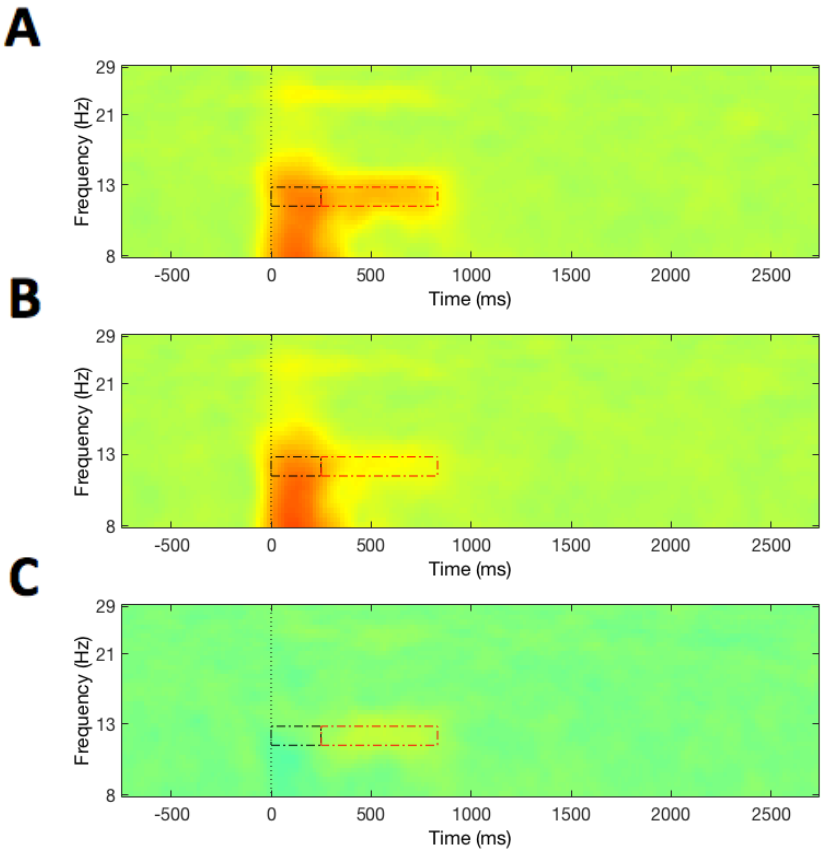


Figure 34.8. ITC analyses of the rhythmic condition recorded from A: electrode C1, B: electrode C2, and C: the subtraction of the electrode C2 from the electrode C1. 0 indicates the time of the first pulse of the train. The black rectangle indicates W1 and the red rectangle indicates W2.

Figure 4.9 shows the average ITC values recorded from electrode C1 and C2 at 12Hz in time, with black lines indicating those time points where values were significantly different. Results showed that the increase in ITC at the frequency of stimulation in C1 is significantly different from C2. Specifically, significant difference ($p<0.05$) included timepoints ranging from 335ms to 843ms.

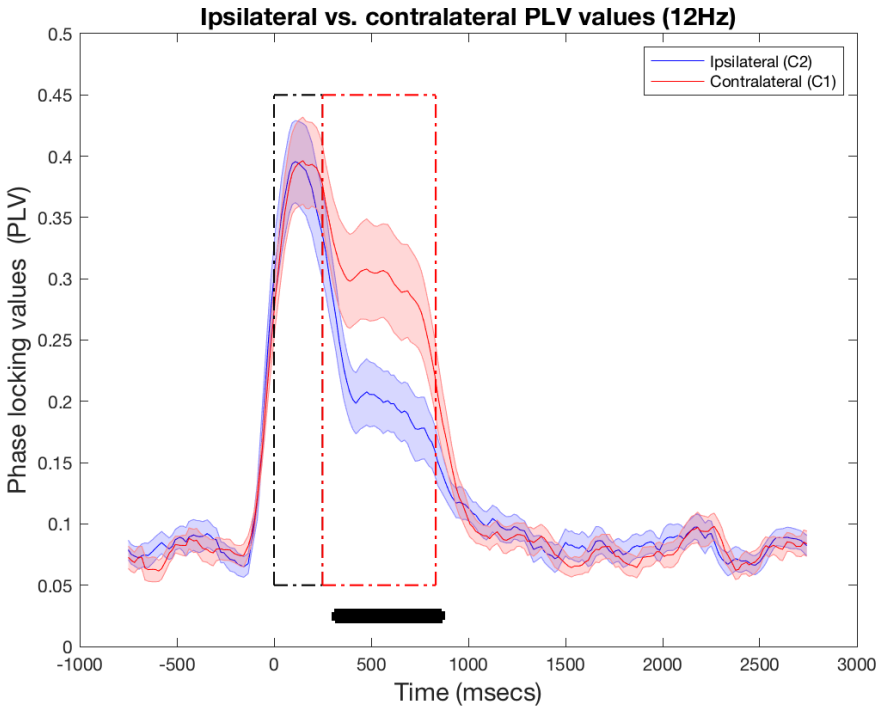


Figure 35.9. ITC values recorded from electrode C1 (red) and electrode C2 (blue) at 12Hz in the rhythmic condition. 0 indicates the time of the first pulse of the train. Black line indicates the time points where the ITC values between electrodes are significantly different. The black rectangle indicates W1 and the red rectangle indicates W2.

Mu waves for each pulse

Figure 4.10 shows the average of mu oscillations at 8-13Hz for each of the ten pulses of the train in both conditions. Results showed a higher amplitude of the oscillations evoked by the first 3 pulses of the train in both conditions, coinciding with both the increase in ERSP power and increase in ITC seen at the beginning of the trains. Also, oscillations in the first 3 pulses of the train show the same phase in both conditions.

After that, during the rest of the pulses (i.e. 4-10 pulses) oscillations show a different phase in both conditions. In rhythmic condition oscillations start oscillating at the same phase after very pulse while in the arrhythmic condition oscillations start oscillations at different phases after very pulse.

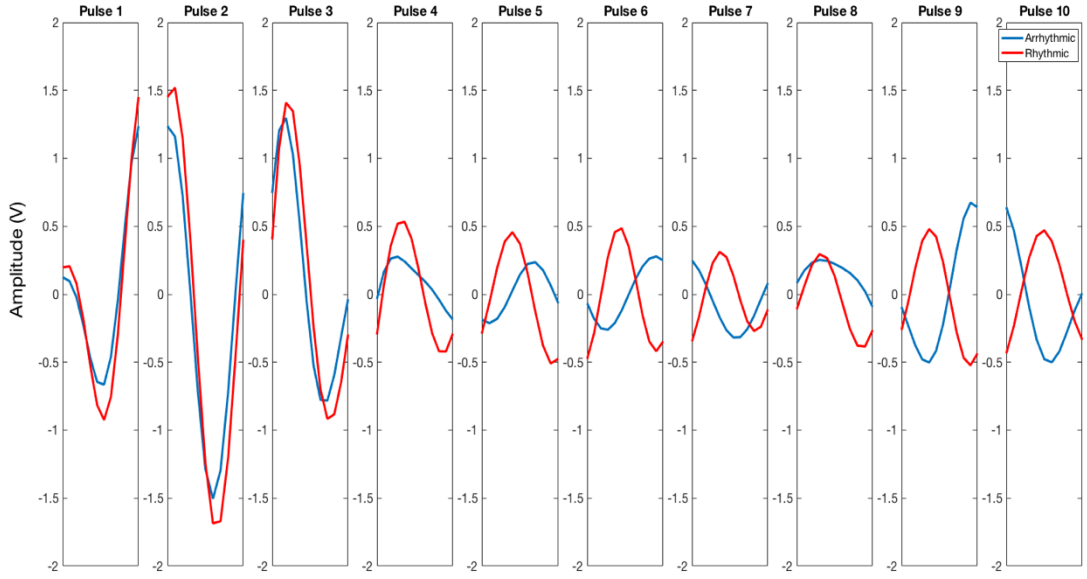


Figure 36.10. Beta oscillations (8-13Hz) for both rhythmic (red) and arrhythmic (blue) conditions for each pulse of the train.

Topography maps

Scalp maps of the ERP were plotted using whole scalp electrodes for the average of pulses 1-3 (0-249ms) and pulses 8-10 (581-832ms) at 8-13Hz in both conditions (see Figure 4.11).

In the average of pulses 1-3 (Figure 4.11A), there was no significant difference between the 64 electrode voltages of both conditions. In the average of pulses 8-10 (Figure 4.11B), both conditions showed a very different scalp map differing significantly ($p<0.05$) in 56 electrode voltages (electrode P7, P9, PO7, O1, Iz, Oz, P10, and O2 were not significantly different).

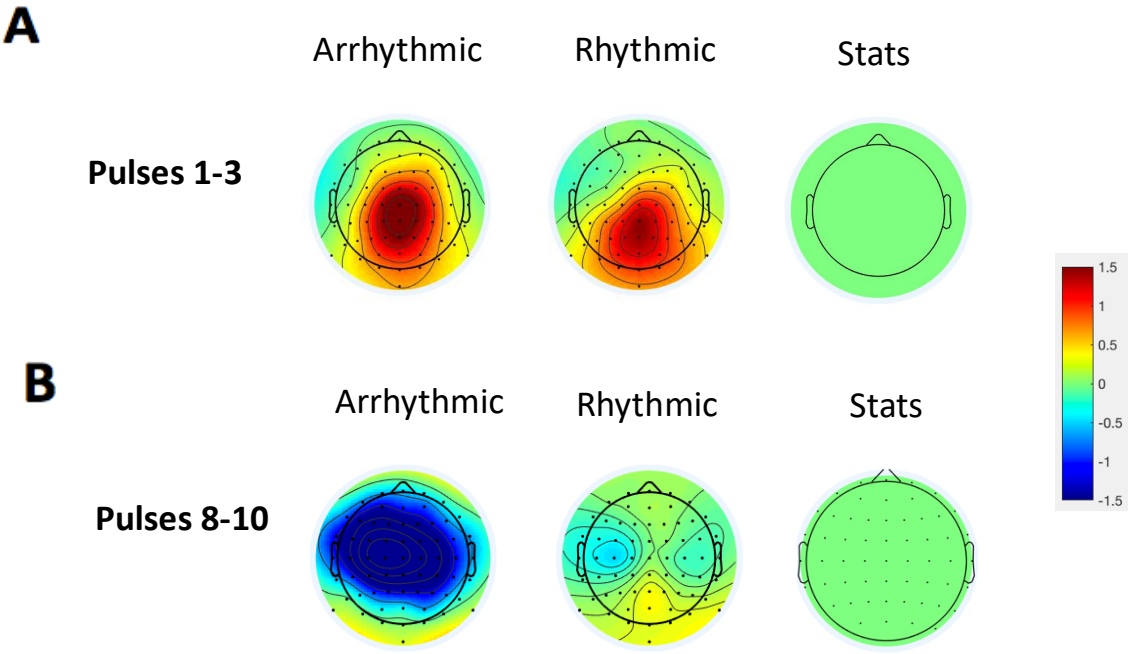


Figure 37.11 Scalp topographies averaged for A: pulses 1-3 and B: pulses 8-10 in the arrhythmic (left) and rhythmic (center) conditions. Scalp maps on the right show the electrodes that showed a significant difference in voltage between conditions.

After-effects of the stimulation on beta band (15-30Hz)

Figure 4.12 shows the time-frequency analyses in both the rhythmic and arrhythmic conditions. Figure 4.13 shows the average ERSP values for the beta (13-30Hz) band for both conditions. Results show that the beta ERD is significantly stronger ($p<0.05$) in the arrhythmic condition compared to the rhythmic condition. Specifically, significant difference ($p<0.05$) included timepoints ranging from 953ms to 1109ms. Results also show that the ERS is significantly stronger ($p<0.05$) in the arrhythmic condition compared to the rhythmic condition. Specifically, significant difference ($p<0.05$) included timepoints ranging from 1335ms to 1953ms.

In the mu band (8-13Hz) there was no significant difference ($p<0.05$) in the ERSP values between conditions after the stimulation.

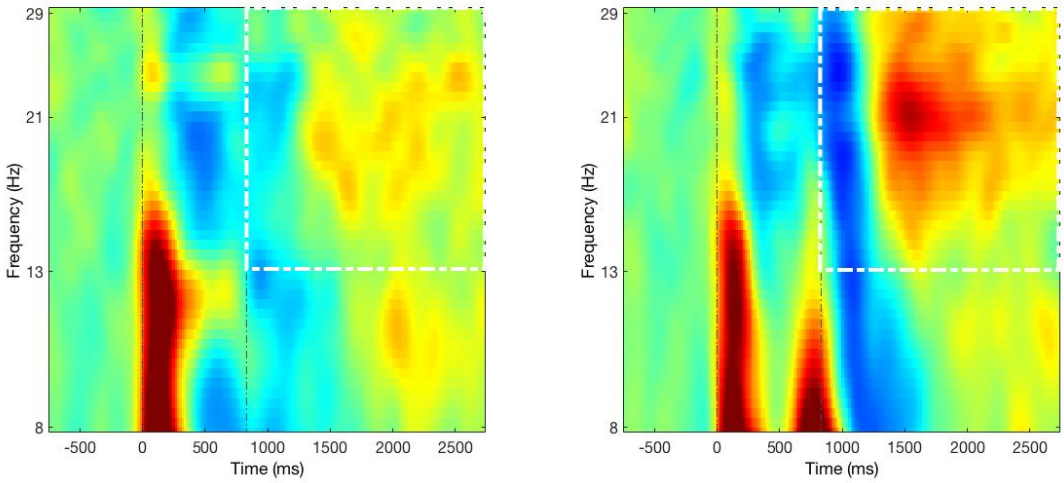


Figure 39.12. Time frequency analyses for electrode C1 for the rhythmic condition (left) and the arrhythmic condition (right). 0 indicates the time of the first pulse of the train. The vertical dotted line indicates the end of the train. The white rectangle indicates beta band (13-30Hz) after the stimulation.

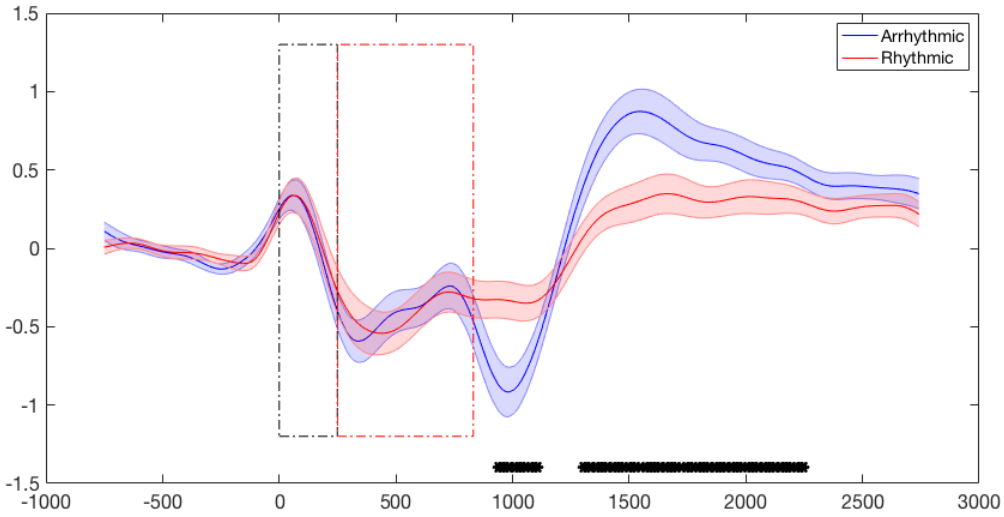


Figure 38.13. Time-frequency values for electrode C1 at the average frequency of 13-30Hz for both the rhythmic (red) and arrhythmic (blue) conditions. 0 indicates the time of the first pulse of the train. Black lines indicate the time points where the time-frequency values between both conditions are significantly different. The black rectangle indicates W1 and the red rectangle indicates W2.

4.3. Beta entrainment study

4.3.1 Methods

Participants

Twenty healthy adult participants (8 males) between 20 and 35 (mean age of 25.2) took part in the study. All of the subjects were right-handed. There were no requirements to take part in the study. Participants were given 8 pounds for their time. None of the recruited participants withdrew from the study. All participants signed the consent form of the study, approved by the Ethics Committee in Nottingham University.

EEG set-up and data collection

EEG set-up and data collection were performed as explained in the General Methods.

Devices

The stimulator was controlled by a bespoke MATLAB vR2017a script on a Mac Pro computer running High Sierra (v 10.13.6).

Experimental procedure

A number of 300 trains of 10 short pulses were delivered every 4 seconds to the right median nerve of the participants, while EEG was being recorded. The trains were delivered in two different conditions randomly. In the rhythmic condition, pulses were delivered every 52ms (i.e. 19Hz); in the arrhythmic condition, pulses were delivered during the same time-window as the rhythmic

condition but in random intervals of time. Figure 4.14 shows a scheme of the design of the study.

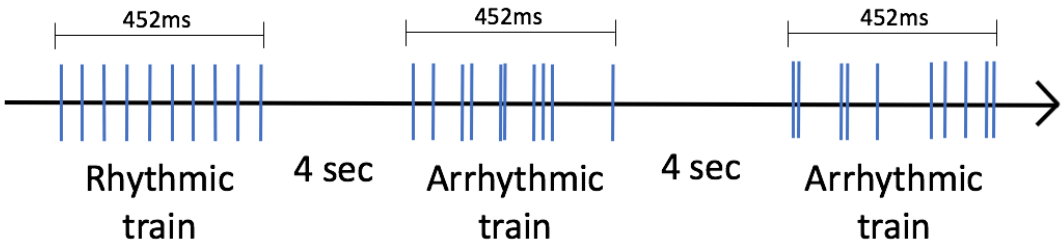


Figure 40.14. Scheme of the design of the study

The duration of a pulse was 200μs and the total length of the train was 452ms. Three short breaks were provided throughout the testing, one every 75 trains. Participants were asked to stay still, relax and keep their eyes open. Participants were monitored with a camera to make sure that the thumb twitch was always present. Intensity was adjusted if needed.

The starting intensity threshold was determined with a trial and error procedure with one pulse until a noticeable thumb twitch was seen.

Pre-processing of the EEG data

Pre-processing of the EEG data was performed as explained in the General Methods. Specifically, data was low-pass filtered at 30Hz and high-pass filtered at 8Hz, which included alpha and beta bands. Time-windows of -1 to 3 seconds, time-locked to the first pulse of the train were extracted.

In total, only one channel was deleted from one participant. The average number of epochs between participants after epoch rejection was 129 in the rhythmic condition and 126 in the arrhythmic condition.

Data Analysis

Data analyses of the EEG data was performed to obtain the ERSP and ITC; beta oscillations (13-30Hz), and scalp maps using whole scalp electrodes.

Statistical analyses were performed using a one-tailed paired-samples t-test correcting for the error of multiple comparisons using False Discovery Rate (FDR; Benjamini & Hochberg, 1995).

4.3.2 Results

All analyses were performed on electrode C1.

Event Related Spectral Perturbation (ERSP)

Time-frequency analyses showed an increase in power at the beginning of the train involving both mu and beta bands in both rhythmic and arrhythmic conditions (see Figure 4.15A-4.15B).

After that, the rhythmic condition showed an increase in beta-power in a narrow band that peaked at the frequency of the stimulation (i.e. 19Hz) (see Figure 4.15B). This increase in power centred at 19Hz was absent in the arrhythmic condition (see Figure 4.15A). This difference becomes clearer when subtracting the arrhythmic condition from the rhythmic condition (see Figure 4.15C).

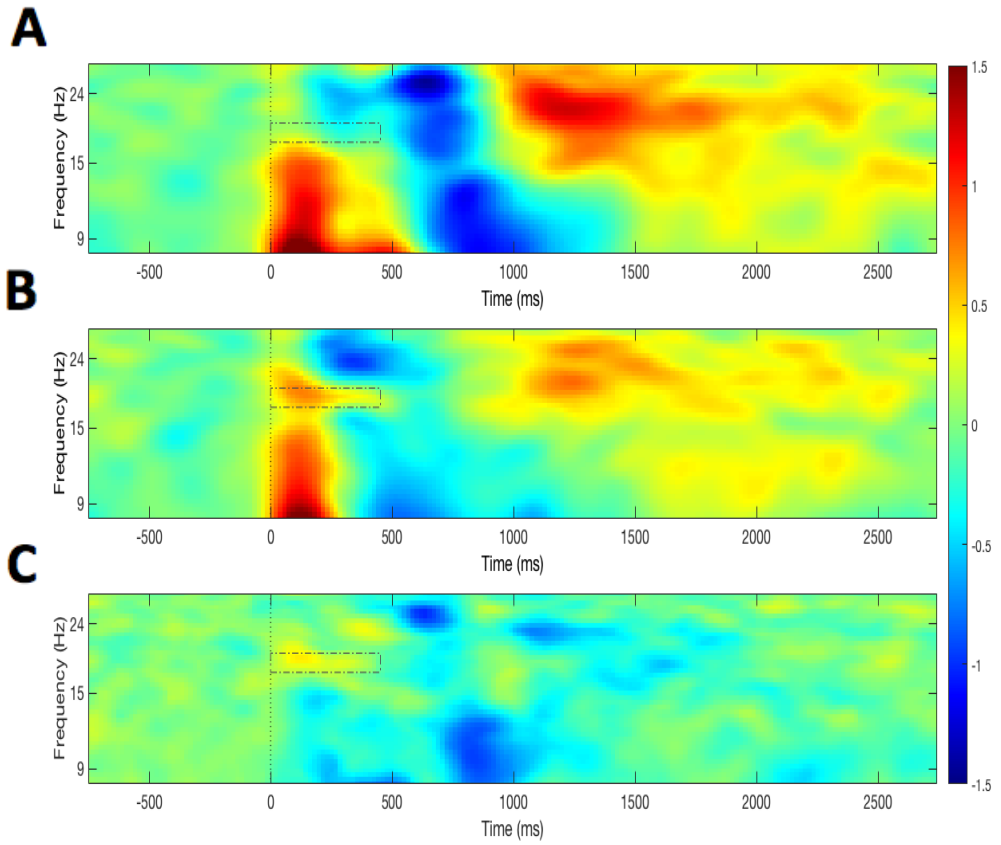


Figure 41.15. Time frequency analyses for electrode C1 for A: the arrhythmic condition, B: the rhythmic condition, and C: the subtraction of the arrhythmic condition from the rhythmic condition. 0 indicates the time of the first pulse of the train. The rectangle indicates the 18-20Hz frequency for the whole length of the train

Figure 4.16 shows the average of ERSP values recorded from electrode C1 at 18-20Hz for both conditions in time, with black lines indicating those time points where values were significantly different. Results showed that the increase in beta power at the frequency of stimulation specific of the rhythmic stimulation is significantly different from the arrhythmic condition. Specifically, significant difference ($p<0.05$) included timepoints ranging from 78ms to 375ms. ERD power was also found to be significantly stronger in the arrhythmic condition compared to the rhythmic condition from 601ms to 898ms.

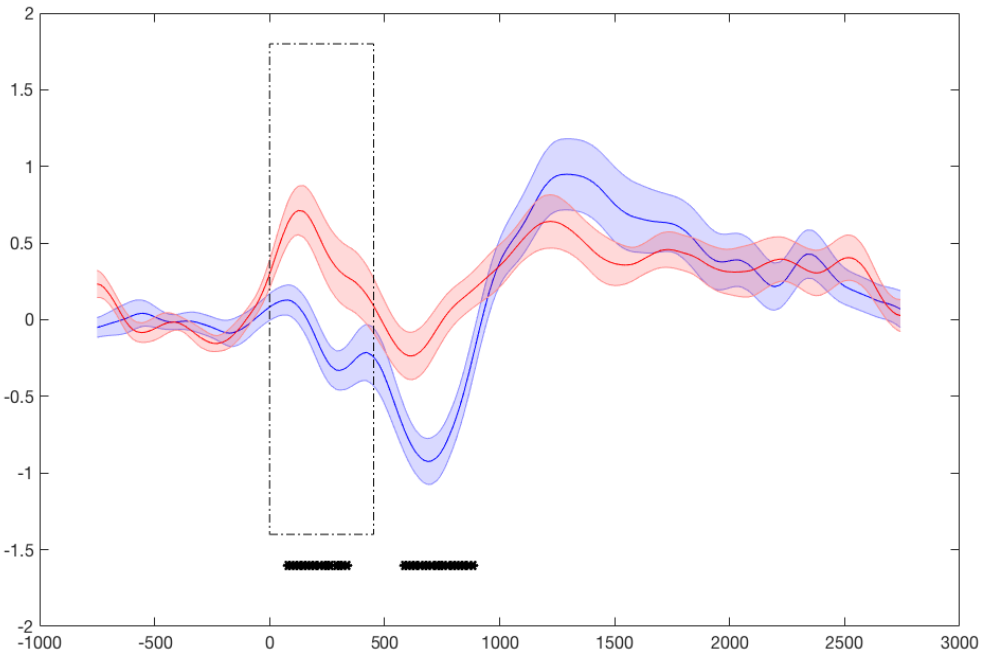


Figure 42.16. Time-frequency values for electrode C1 at the average frequency of 18-20Hz for both the rhythmic (red) and arrhythmic (blue) conditions. 0 indicates the time of the first pulse of the train. Black line indicates the time points where the time-frequency values between both conditions are significantly different. The rectangle indicates the length of the train

Inter-trial coherence (ITC)

ITC analyses showed an increase in phase locking at the beginning of the train involving both mu and beta bands in both rhythmic and arrhythmic conditions (see Figure 4.17A-4.17B).

After that, the rhythmic condition showed an increase in ITC in a narrow band that peaked at the frequency of the stimulation (i.e. 19Hz) (see Figure 4.17B). This increase in phase locking centred at 19Hz was absent in the arrhythmic condition (see Figure 4.17A). This difference becomes clearer when subtracting the arrhythmic condition from the rhythmic condition (see Figure 4.17C).

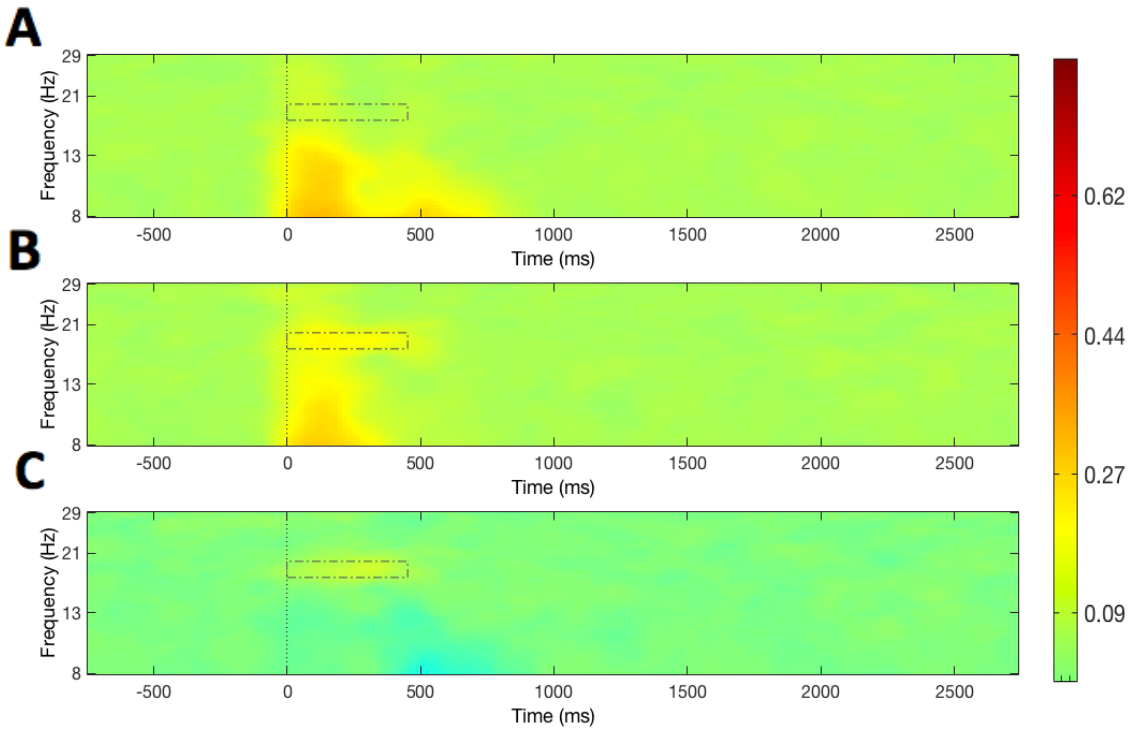


Figure 43.17. ITC analyses for electrode C1 for A: the arrhythmic condition, B: the rhythmic condition, and C: the subtraction of the arrhythmic condition from the rhythmic condition. 0 indicates the time of the first pulse of the train. The rectangle indicates the 18-20Hz frequency for the whole length of the train

Figure 4.18 shows the average of ITC values recorded from electrode C1 at 18-20Hz for both conditions in time, with black lines indicating those time points where values were significantly different. Results showed that the increase in ITC at the frequency of stimulation specific of the rhythmic stimulation is significantly different from the arrhythmic condition. Specifically, significant difference ($p<0.05$) included timepoints ranging from 54ms to 460ms.

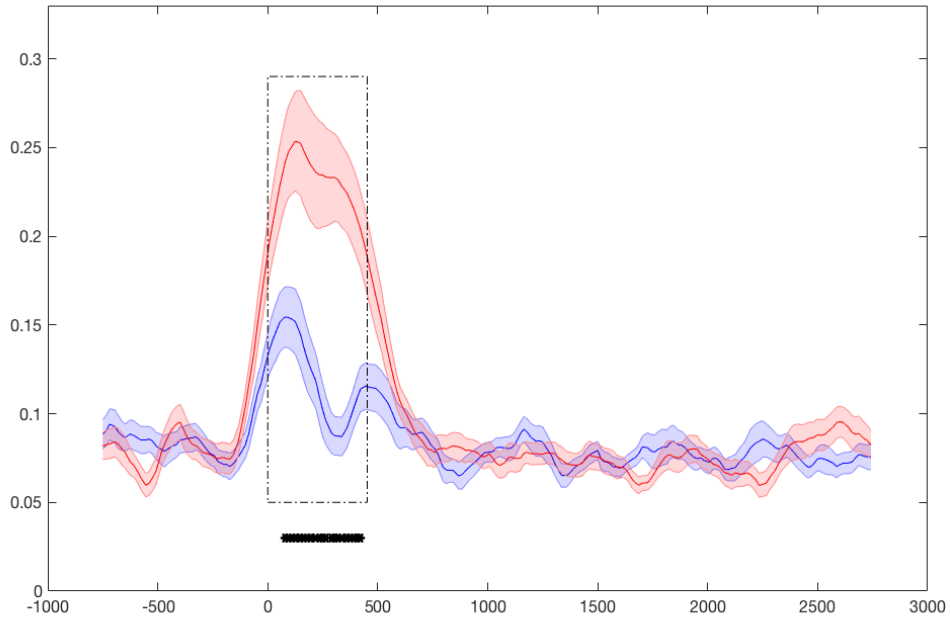


Figure 44.18. ITC values for electrode C1 at the average frequency of 18-20Hz for both the rhythmic (red) and arrhythmic (blue) conditions. 0 indicates the time of the first pulse of the train. Black line indicates the time points where the ITC values between both conditions are significantly different. The rectangle indicates the whole length of the train

Inter-hemisphere effect

Figure 4.19A-4.19B shows the time-frequency analyses in the rhythmic condition from electrode C1 and C2. Electrode C1 is placed over the contralateral hemisphere to the stimulation while electrode C2 is placed over the ipsilateral hemisphere to the stimulation.

Results showed an increase in power at the beginning of the train involving both mu and beta bands in both C1 and C2 electrodes (see Figure 4.19A-4.19B). After that, the contralateral hemisphere showed an increase in beta-power in a narrow band that peaked at the frequency of the stimulation (i.e. 19Hz) (see Figure 4.19A). This increase in power centred at 19Hz was absent in the ipsilateral hemisphere (see Figure 4.19B). This difference becomes clearer when subtracting C2 from C1 (see Figure 4.19C).

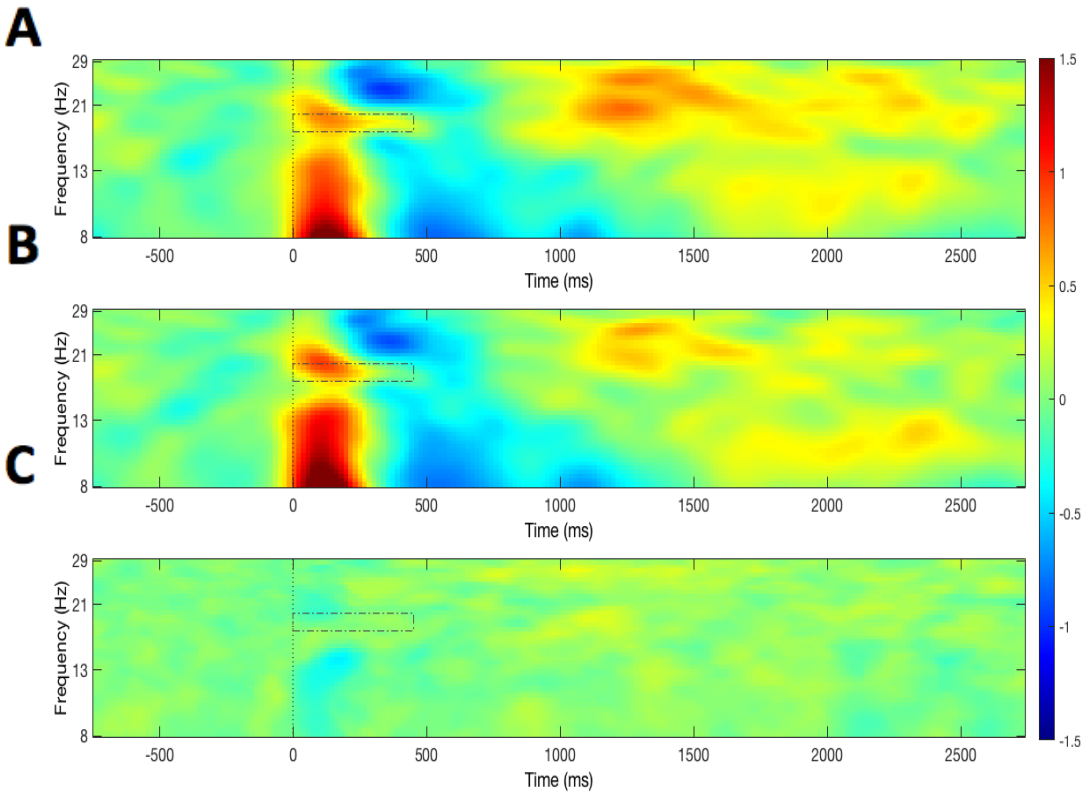


Figure 45.19. Time frequency analyses of the rhythmic condition for A: electrode C1, B: electrode C2, and C: the subtraction of the electrode C2 from the electrode C1. 0 indicates the time of the first pulse of the train. The rectangle indicates the 18-20Hz frequency for the whole length of the train

Figure 4.20 shows the average ERSP values recorded from electrode C1 and C2 at 18-20Hz in time, with black lines indicating those time points where values were significantly different. Results showed that the increase in beta power at the frequency of stimulation in C1 is significantly different from C2. Specifically, significant difference ($p<0.05$) included timepoints ranging from 406ms to 562ms.

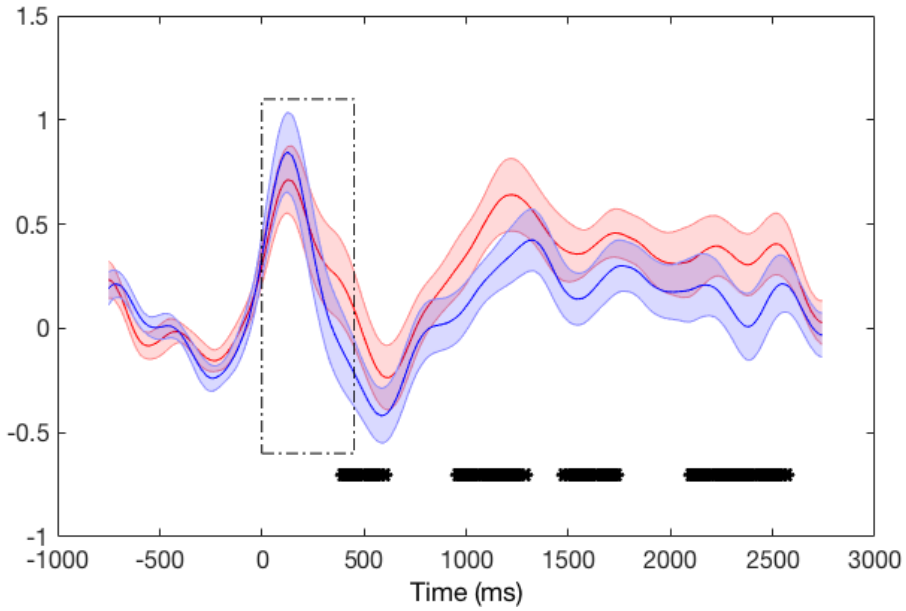


Figure 46.20. Time-frequency values for electrode C1 (red) and electrode C2 (blue) at the average frequency of 18-20Hz for the rhythmic conditions. 0 indicates the time of the first pulse of the train. Black line indicates the time points where the time-frequency values between both electrodes are significantly different. The rectangle indicates the whole length of the train

Moreover, beta rebound in the ipsilateral hemisphere show significantly stronger ERS than the contralateral hemisphere from 984ms to 1265ms, from 1476ms to 1687ms and from 2093ms to 2531ms.

Figure 4.21A-4.21B shows the ITC analyses in the rhythmic condition from electrode C1 and C2. Electrode C1 is placed over the contralateral hemisphere to the stimulation while electrode C2 is placed over the ipsilateral hemisphere to the stimulation.

Results showed an increase in phase locking at the beginning of the train involving both mu and beta bands in both C1 and C2 electrodes (see Figure 4.21A-4.21B). After that, the contralateral hemisphere showed an increase in ITC in a narrow band that peaked at the frequency of the stimulation (i.e. 19Hz) (see Figure 4.21A). This increase in power centred at 19Hz was absent in the ipsilateral hemisphere (see Figure 4.21B). This difference becomes clearer when subtracting C2 from C1 (see Figure 4.21C).

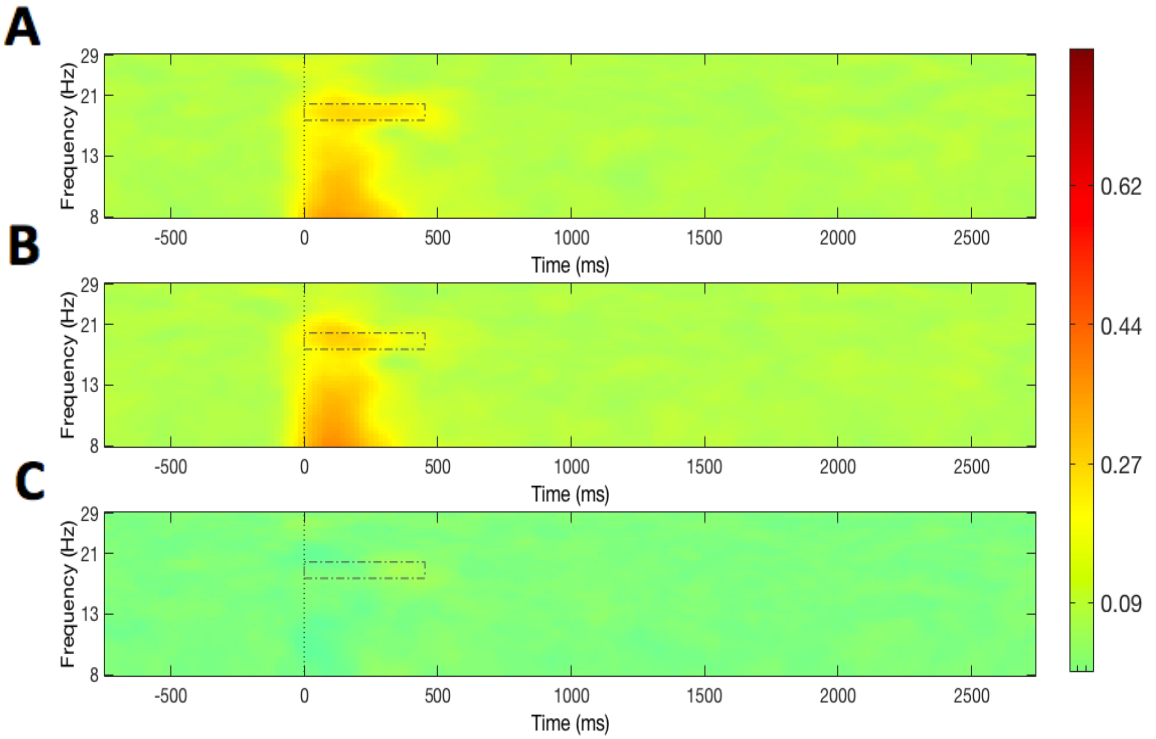


Figure 47.21. Inter-trial coherence analyses of the rhythmic condition for A: electrode C1, B: electrode C2, and C: the subtraction of the electrode C2 from the electrode C1. 0 indicates the time of the first pulse of the train. The rectangle indicates the 18-20Hz frequency for the whole length of the train

Figure 4.22 shows the average ITC values recorded from electrode C1 and C2 at 18-20Hz in time, with black lines indicating those time points where values were significantly different. Results showed that the increase in ITC at the frequency of stimulation in C1 is significantly different from C2. Specifically, significant difference ($p<0.05$) included timepoints ranging from 375ms to 421ms.

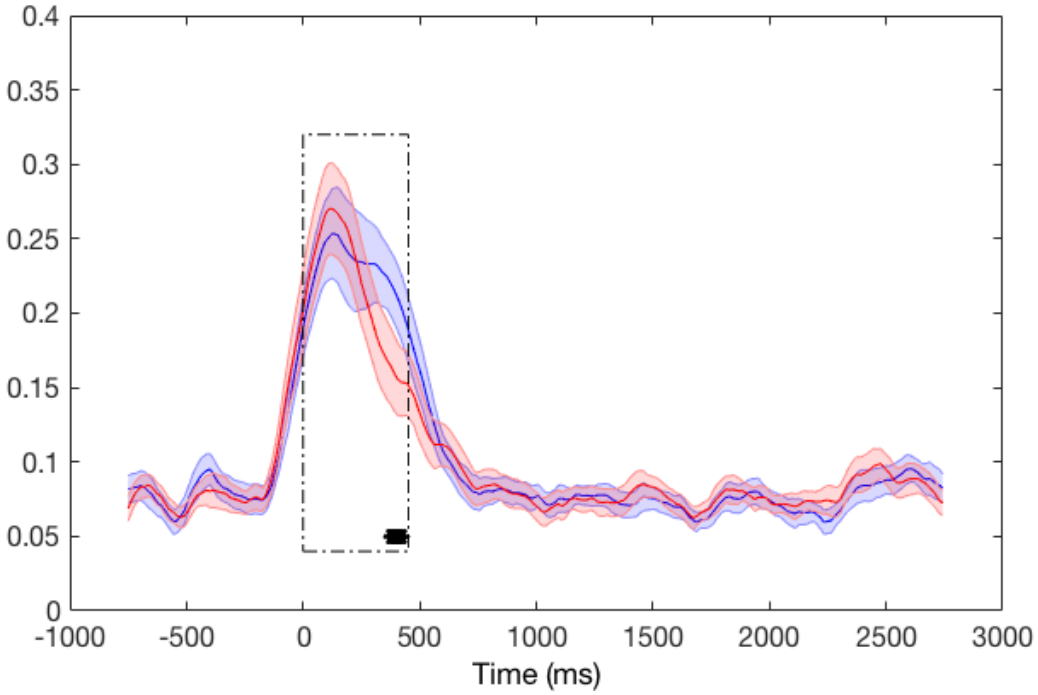


Figure 48.22. ITC values for electrode C1 (red) and electrode C2 (blue) at the average frequency of 18-20Hz for the rhythmic condition. 0 indicates the time of the first pulse of the train. Black line indicates the time points where the ITC values between electrodes are significantly different. The rectangle indicates the whole length of the train

Beta waves for each pulse

Figure 4.23 shows the average of beta oscillations at 13-30Hz for each of the ten pulses of the train in both conditions. Results showed a higher amplitude of the oscillations evoked by the first 4 pulses of the train in both conditions, coinciding with both the increase in ERSP power and increase in ITC seen at the beginning of the train. Also, oscillations in the first 4 pulses of the train show the same phase in both conditions. After that, during the rest of the pulses (i.e. 5-10 pulses) oscillations show a different phase in both conditions.

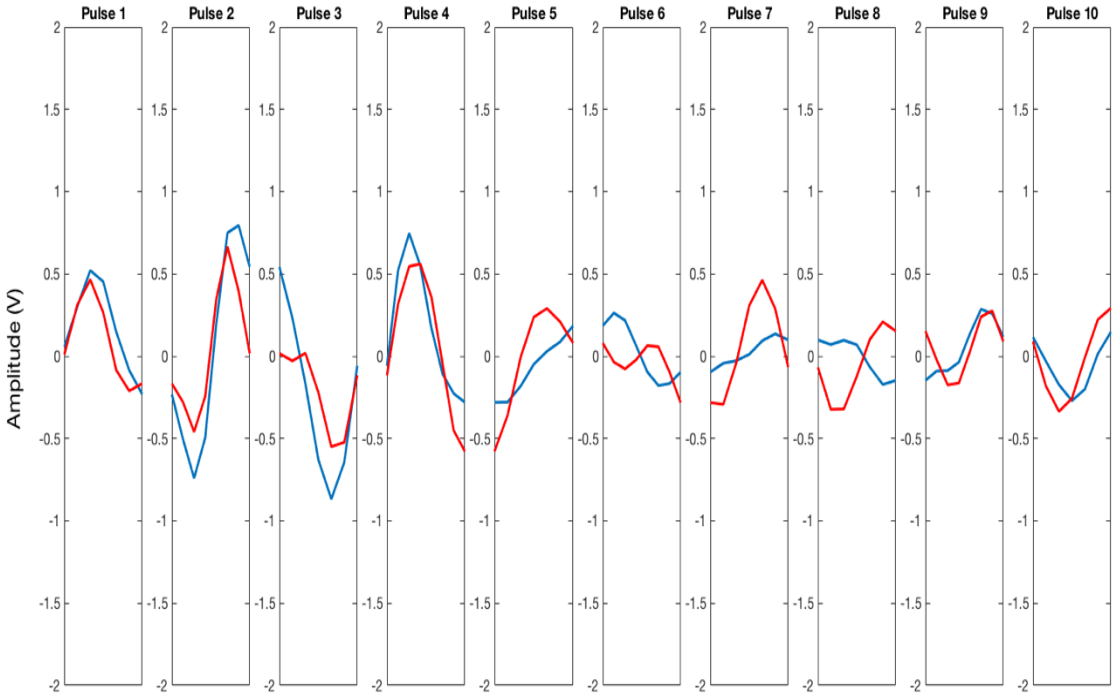


Figure 49.23. Beta oscillations (13-30Hz) for both rhythmic (red) and arrhythmic (blue) conditions for each pulse of the train.

Topography maps

Scalp maps of the ERP were plotted using whole scalp electrodes for the average pulses 1-3 (0-150ms) and pulses 8-10 (350-500ms) at 13-30Hz in both conditions (see Figure 4.24).

In the average of pulses 1-3 (Figure 4.24A), there was no significant difference between the 64 electrode voltages of both conditions. In the average pulses 8-10 (Figure 4.24B), both conditions show a different scalp map differing significantly ($p<0.05$) in 57 electrode voltages (electrodes P7, P9, P07, O1, Iz, P10, TP7 were not significantly different).

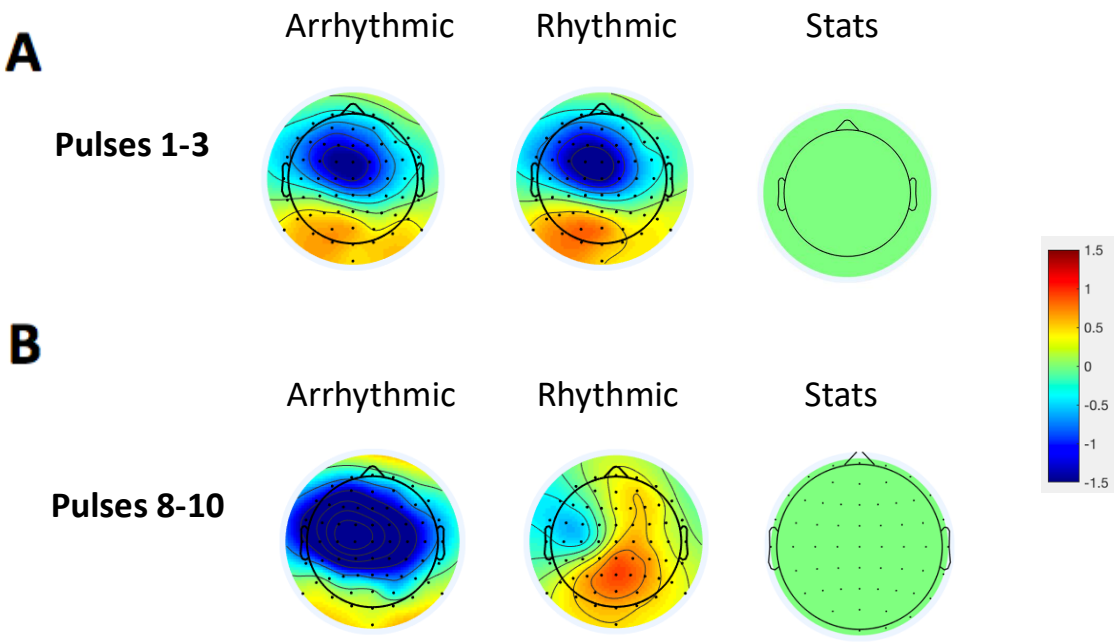


Figure 50.24. Scalp topographies averaged for A: pulses 1-3 and B: pulses 8-10 in the arrhythmic (left) and rhythmic (center) conditions. Scalp maps on the right show the electrodes that showed a significant difference in voltage between conditions.

After-effects of the stimulation

Figure 4.25 shows the time-frequency analyses in both the rhythmic and arrhythmic conditions. Figure 4.26 shows the average ERSP values for the mu (8-13Hz) band for both conditions. Results show that the mu ERD is significantly ($p<0.05$) stronger in the arrhythmic condition compared to the rhythmic condition. Specifically, significant difference ($p<0.05$) included timepoints ranging from 773ms to 914ms. Mu ERS was not significantly different.

Figure 4.27 shows the average ERSP values for the high beta band (21-30Hz) for both conditions. Results showed that the high-beta ERD and ERS are significantly stronger in the arrhythmic condition compared to the rhythmic condition. Specifically, significant ERD difference ($p<0.05$) included timepoints

ranging from 585ms to 898ms; and significant ERS difference ($p<0.05$) included timepoints ranging from 1023ms to 1195ms and from 2359ms to 2460ms.

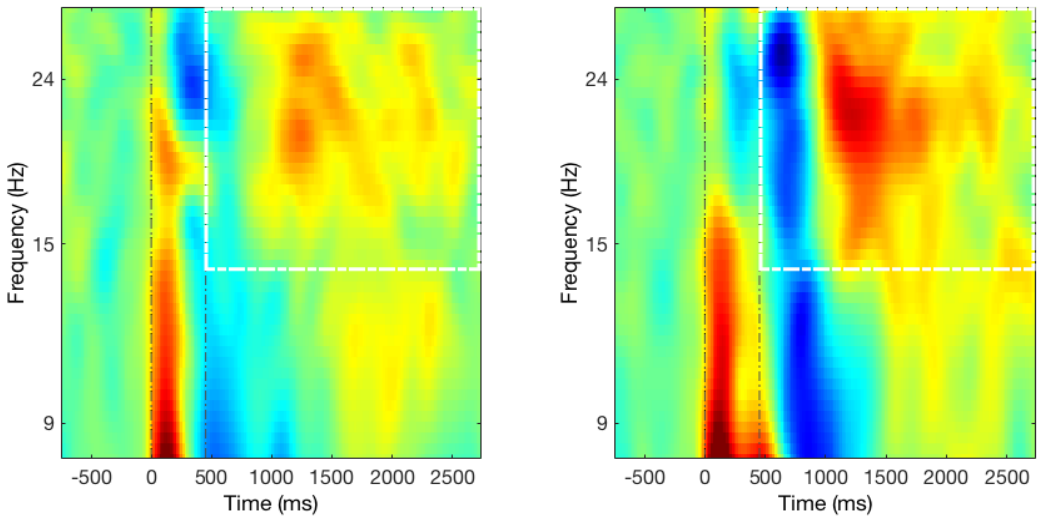


Figure 51.25. Time frequency analyses for electrode C1 for the rhythmic condition (left) and the arrhythmic condition (right). 0 indicates the time of the first pulse of the train. The vertical dotted line indicates the end of the train

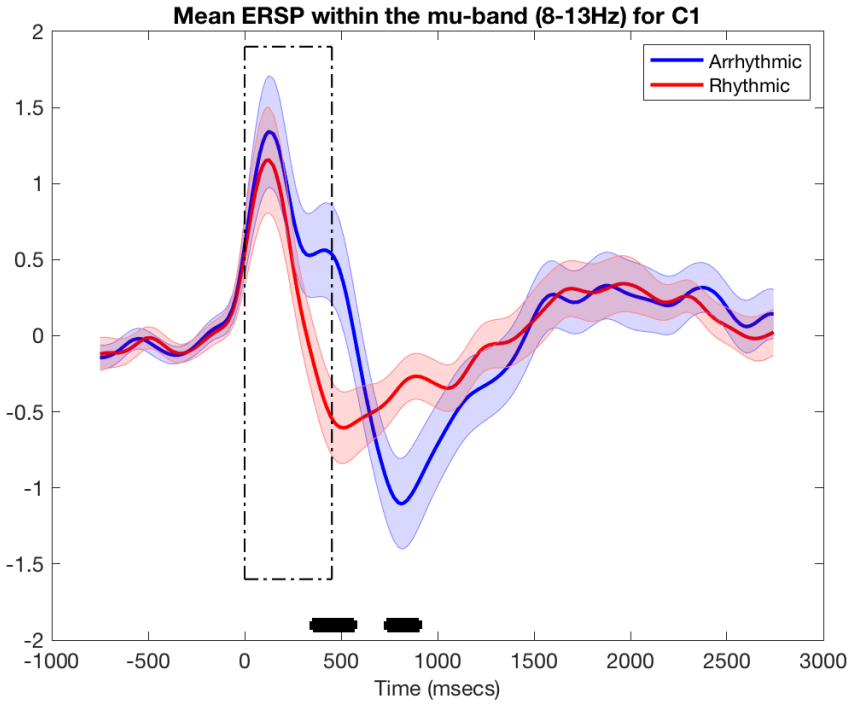


Figure 52.26. Time-frequency values for electrode C1 at the average frequency of mu (8-13Hz) for both the rhythmic (red) and arrhythmic (blue) conditions. 0 indicates the time of the first pulse of the train. Black line indicates the time points where the time frequency values between both conditions are significantly different. The rectangle indicates the whole length of the train

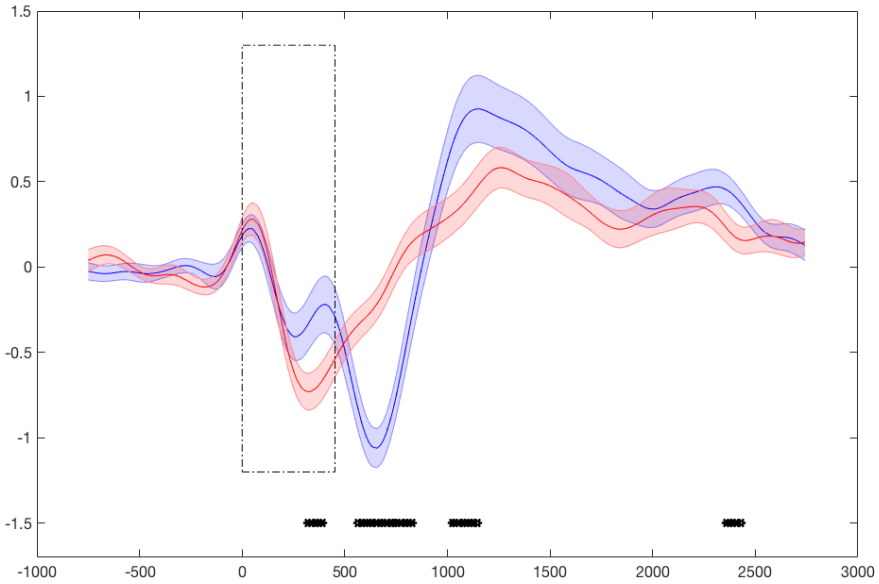


Figure 53.27. Time-frequency values for electrode C1 at the average frequency of 21-30Hz for both the rhythmic (red) and arrhythmic (blue) conditions. 0 indicates the time of the first pulse of the train. Black line indicates the time points where the time-frequency values between both conditions are significantly different. The rectangle indicates the whole length of the train

4.4. Discussion

ERSP and ITC

The main finding is that rhythmic stimulation delivered with MNS results in the entrainment signature exposed by Thut et al (2011), characterized by an initial increase in EEG power involving both mu and beta frequency bands followed by an increase in EEG power at the frequency of stimulation of the target area. Specifically, results show that 12Hz rhythmic stimulation and 19Hz rhythmic stimulation delivered with MNS on the right wrist results in an increase in EEG power centred at 12Hz and 19Hz at the contralateral motor cortex, respectively.

Initial increased ITC involving both mu and beta frequency bands followed by an increase in ITC at the frequency of stimulation of the target area corroborates that the increase in power is due to an increase in phase synchronization of the oscillations from the targeted neural populations. Specifically, results show that

12Hz rhythmic stimulation and 19Hz rhythmic stimulation delivered with MNS on the right wrist results in an increase in ITC centred at 12Hz and 19Hz at the contralateral motor cortex, respectively.

The initial increase in EEG power and ITC was also present when the MNS was delivered arrhythmically. However, the following increase in EEG power and ITC seen in the rhythmic condition was absent in the arrhythmic condition. Specifically, the delivery of random pulses at an average of 12Hz and 19Hz frequency showed no increase in power or ITC in the target area at 12Hz or 19Hz, respectively. This absence of increased ITC at the frequency of stimulation corroborates that the arrhythmic stimulation did not result in phase synchronization of the oscillations from the targeted neural populations at the frequency of stimulation. Moreover, the absence of both increased EEG power and ITC at the frequency of stimulation suggests that there is no increased power without increased ITC.

The fact that the initial increase in EEG power and ITC was seen in both rhythmic and arrhythmic conditions is evidence that it corresponds to the mere response to receiving stimulation, independently of what type of stimulation is being delivered. The fact that the following effect was only seen in the rhythmic condition is evidence that it corresponds to the specific type of stimulation delivered.

An initial broad increase in power over both mu and beta followed by an increase in power over the frequency of stimulation was also seen when delivering a train of 5 TMS pulses (Thut et al., 2011) and can be similarly explained by the initial pulses of the stimulation causing an arousal effect on neural populations oscillating at many different frequencies and the rest of the pulses of the stimulation recruiting only those populations oscillating at the frequency of stimulation to phase align their oscillations to the rhythmic pulses.

Ipsilateral vs contralateral

Both the initial increase in power and initial increase in ITC was seen in both the contralateral and ipsilateral hemisphere when stimulating at 12Hz and when stimulating at 19Hz. The increase in EEG power and ITC at the frequency of stimulation was seen in the contralateral cortex but not in the ipsilateral cortex when stimulating at 12Hz and when stimulating at 19Hz.

These results show that the initial pulses create a general effect that occurs in both hemispheres, but the following effect is more specific to the contralateral hemisphere to the stimulation.

Brain oscillations

Mu oscillations (8-13Hz), when stimulating at 12Hz, and beta oscillations (13-30Hz), when stimulating at 19Hz, show two different patterns of behaviour during the 10 pulses of the train in both the rhythmic and arrhythmic conditions. Initially, both rhythmic and arrhythmic conditions show an increased amplitude of the oscillations as well as an initial synchronization in phase between conditions. This is seen for the first 3 pulses in the rhythmic (12Hz) condition and for the first 4 pulses in the rhythmic (19Hz) condition. The fact that the initial pulses evoke a higher amplitude is evidence that the stimulation is producing a response on many different neural populations, and the fact that both conditions oscillate at the same phase is evidence that the stimulation is causing a reset in phase of the oscillations from those affected neural populations.

After these pulses, both conditions oscillate at different phases. When stimulating at 12Hz, oscillations in the rhythmic condition oscillate at the same phase after every pulse while oscillations in the arrhythmic condition oscillate at different phases after every pulse. When stimulating at 19Hz, in the three last pulses of the rhythmic condition, oscillations start their oscillation at the same phase, while in the arrhythmic condition, oscillations start oscillating at a different phase for all of the six pulses. This suggests that the rhythmic

stimulation is resetting the phase from those neural populations that the stimulation is recruiting to oscillate at the frequency of stimulation.

These results also suggest that the increase in EEG power and ITC produced by the initial pulses of the train is due to both a higher amplitude of the oscillations and phase synchronization, while the increase at the frequency of stimulation is merely due to phase synchronization. This explains the difference in EEG power in the rhythmic condition between the initial pulses and the rest of the train.

Topographies

Average of whole-scalp maps for the first 3 pulses of the train show that, independently of which frequency is being stimulated, the initial pulses of the train evoke a response that is found in a similar location in both conditions, while the last 3 pulses of the train evoke a response that is found in a different location in each condition. In the arrhythmic condition, potentials are widespread throughout the scalp, while in the rhythmic condition, potentials are more local to the contralateral motor cortex. This is consistent with the fact that the lasts of the rhythmic electric pulses evoke a response only to the neural populations being recruited by the stimulation in the motor cortex, while the arrhythmic electric pulses have a more general effect.

Aftereffects

As previously reported, MNS leads to a reduction in power in both mu and beta bands, followed by an increase in power in both mu and beta bands.

Interestingly, rhythmic stimulation had an effect on these later responses to the stimulation. When stimulating the mu band (i.e. 12Hz), the beta ERD and beta ERS showed a significantly reduced power in the rhythmic condition compared to the arrhythmic condition. When stimulating the beta band (i.e. 19Hz) both the

mu ERD and high beta ERD, as well as the high beta ERS, showed a significantly reduced power in the rhythmic condition compared to the arrhythmic condition.

This suggests that, rhythmic stimulation, independently of which frequency band is being stimulated, has an effect on the beta band by reducing the power of the beta ERD and beta ERS. Rhythmic stimulation at the beta band has also an effect on mu band by reducing the power of the mu ERD. This reduced power found in the rhythmic conditions could be explained by the fact that the end of the rhythmic stimulation train affects a lower amount of neural populations compared to the end of the arrhythmic stimulation train, where a second broad increase in power is produced. This is supported by Houdayer (2006) suggestion that the magnitude of beta ERS is stronger when the number of afferent inputs is higher.

The ipsilateral hemisphere showed a significantly reduced beta ERS compared to the contralateral hemisphere, independently of which frequency band was being stimulated. Since the beta ERS in the ipsilateral hemisphere is solely produced by the stimulation while the beta ERS in the contralateral hemisphere is due to both the stimulation and the finger movement produced by the stimulation, this again supports Houdayer (2006) suggestion that the magnitude of beta ERS is stronger when the number of afferent inputs is higher.

4.5. Conclusion and continuance

These two studies showed that the delivery of rhythmic MNS leads to the entrainment of the targeted brain oscillations in the contralateral motor cortex, characterized by an increase in power and ITC at the frequency of stimulation.

These findings provide with a new method to modulate brain oscillations in the motor cortex involving a technique that, as opposed to current used techniques (TMS, tES), does not require screening and is easily applied.

The following chapter will explore the effects of rhythmic MNS on motor behaviour in HS.

Note - The Mu entrainment study (4.2) was included in the published paper:

Maiquez, B. M., Sigurdsson, H. P., Dyke, K., Clarke, E., McGrath, P., Pasche, M., ... & Jackson, S. R. (2020). Entraining Movement-Related Brain Oscillations to Suppress Tics in Tourette Syndrome. *Current Biology*

CHAPTER 5

Chapter 5 - Exploring MNS behavioural effects of mu entrainment

---

5.1. Introduction

It is known that brain oscillations play a role in cognitive functions. Specifically, mu and beta oscillations in the SMC are involved in movement control, were the desynchronization of mu and beta oscillations is related to movement preparation and movement execution, respectively, while the synchronization of beta oscillations is related to movement termination (G Pfurtscheller & Lopes, 1999).

Entrainment increases the synchronization of brain oscillations resulting in an increase of cortical inhibition. Depending on which cortical region of the brain the entrainment is applied to, this will have a behavioural effect specific to the cognitive function modulated by that region. For example, the entrainment of beta oscillations in the motor cortex has shown to reduce force (Joundi et al., 2012) and slow down velocity in voluntary movement (Pogosyan et al., 2009).

Summary and aim

In the last chapter, I have shown that entrainment of mu oscillations in the SMC using MNS reduces motor cortical excitability. The aim of this study is to investigate whether entrainment of mu oscillations in the motor cortex using MNS has a behavioural effect in motor responses.

Mu ERD is involved in movement preparation (Leocani, Toro, Manganotti, Zhuang, & Hallett, 1997; G Pfurtscheller, 1989), and as shown in Chapter 3a.1, it starts before the initiation of the movement. I expect that by entraining the

motor cortex I will be increasing the amount of desynchronization needed to execute the movement, delaying the start of the movement initiation. I hypothesize that entrainment of mu oscillations in the SMC using MNS will delay motor initiation.

5.2. Methods

Participants

In Study 1, 20 healthy adult participants (6 males) between 18 and 25 (mean age of 20.8) took part in the study. In Study 2, 20 healthy adult participants (6 females) between 18 and 25 (mean age of 20.6) took part in the study. None of the subjects were left-handed. None of the 40 recruited participants withdrew from the study. All participants signed the consent form of the study, approved by the Ethics Committee in Nottingham University.

Stimulation

Trains of 8 short pulses were delivered to the right median nerve of the participants lasting a total of 582.6ms. The trains were delivered in two different conditions. In the rhythmic condition, pulses were delivered every 83ms (i.e. 12Hz); in the arrhythmic condition, pulses were delivered during the same time-window as the rhythmic condition but in random intervals of time. No inter-pulse interval was less than 10ms in the arrhythmic condition. The duration of a pulse was 200μs and the total length of the train was 582.6ms. The intensity of the stimulation used was the minimum intensity at which a thumb twitch was seen.

Design

A fixation cross appeared for 550ms, after the first 300ms, there was 250ms of pre-stimulus stimulation, which involved the delivery of 3 pulses in both the rhythmic and arrhythmic conditions; this was followed by the presentation of either a blue or yellow circle (diameter of 11cm) while the rest of the pulses were being delivered; after that, participants had to make a response as fast as possible. If no response was made, the visual stimuli disappeared after 3 seconds. The response consisted in releasing the left or right arrow buttons in a keyboard, which were being pressed by the index and middle fingers of the right hand. Trials were separated by 4 seconds. Figure 5.1 shows a scheme of the design of the study.

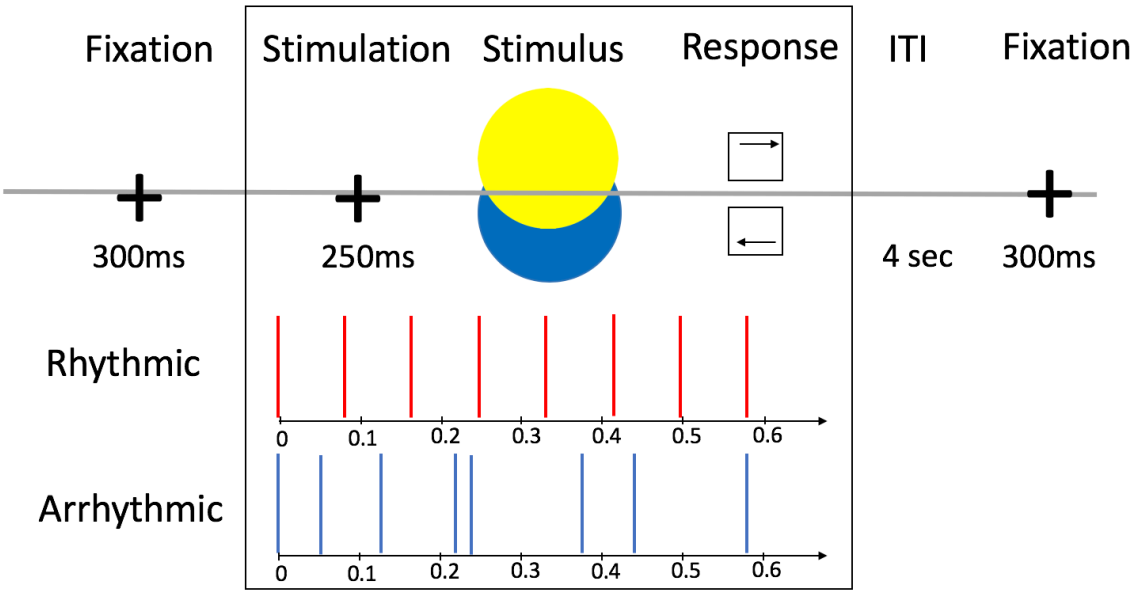


Figure 5455.1. Scheme of the design of the study

Participants were asked to respond as fast as possible. There were 4 blocks, 75 trials per block. There was a total of 300 trials, in 150 of the trials a yellow circle appeared requiring a response from the index finger, in the other 150 trials, a blue circle appeared requiring a response from the middle finger. Each index and middle responses received the same amount of rhythmic and arrhythmic condition.

In study 1, the task was set in a blocked design (ABBA), where in each block the participant received either rhythmic or arrhythmic stimulation; in study 2, the task was set in a random design, where in each block the participant received random trials of rhythmic or arrhythmic stimulation with the same probability.

Devices

The visual stimuli were presented on a computer screen (LG Flatron, screen resolution: 1680 x 1050). Participants made responses on an Apple iMac (A1243) keyboard connected to a MacBook laptop running Windows 8 via USB cable. The task and the stimulator were controlled by a bespoke MATLAB vR2017a script coded using *Psychtoolbox* (http://psychtoolbox.org) on the MacBook.

Experimental procedure

Participants were seated in a small room, approximately 70cm away from the screen and with their right hand resting on a comfortable folded blanket.

The starting intensity threshold was determined with a trial and error procedure with one pulse until a noticeable thumb twitch was seen. This threshold was further validated with a train of 8 pulses at 12 Hz frequency while the participants pressed down on both arrow keys with their right index and middle fingers. Intensity was adjusted if needed.

Then participants proceeded to complete 10 practice trials of the task without any stimulation and 10 practice trials with the stimulation. Prior to the main task and between blocks, the stimulation intensity was re-evaluated with a train of 8 pulses at 12 Hz frequency while the participants pressed down on both arrow keys with their right index and middle fingers and adjusted if needed (either increased or decreased).

Processing of the data

Trials where both buttons were not pressed down simultaneously were rejected; missed trials were rejected; incorrect trials were rejected. Response times (RT) were calculated from the time of the appearance of the stimulus to the time of the release of the arrow key on the keyboard. All RT with a bigger or lower than the average plus standard deviation of 2.5 were rejected to avoid outliers. Average RT were calculated for rhythmic and arrhythmic conditions.

5.3. Results

Study 1: Blocked design

Response Times (RT)

Mean RT for the rhythmic condition was 0.346s (SD 0.027) and the mean RT for the arrhythmic condition was 0.340s (SD 0.026). The difference in RT between rhythmic and arrhythmic conditions was significant (p= 0.029). Figure 5.2 shows boxplots for the RT in each condition. Mean RT corroborate that participants were still receiving stimulation while they were making a response.

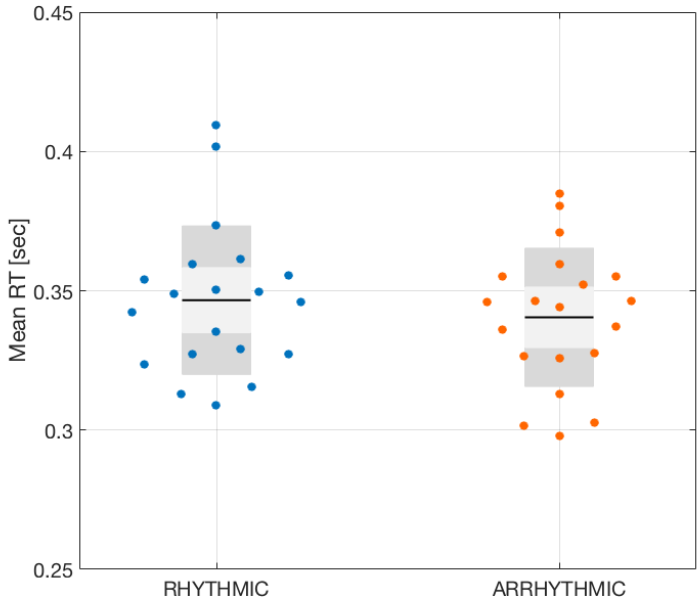


Figure 56.57. Boxplot of the RT for each condition. Every dot represents one subject's average RT

Incorrect trials

In the rhythmic condition, the mean number of incorrect trials was 12.70 (SD 10.34) and in the arrhythmic condition, the mean number of incorrect trials was 12.60 (SD 9.32). This difference was not significant (t(19)=0.0954, p= 0.925).

RT including correct and incorrect trials were rank-ordered in an ascendant order and grouped in 7 bins of 20 trials in each condition. The mean RT and the percentage of incorrect trials was calculated for each bin. Figure 5.3 shows a plot with the mean RT of each bin and the corresponding error percentage for both rhythmic and arrhythmic trials. A quadratic polynomial curve has been fitted to the data.

In the rhythmic condition, the bin range goes from 0.247s to 0.420s, while in the arrhythmic condition, the bin range goes from 0.237s to 0.329s. The fastest bin belongs to the arrhythmic condition while the slowest bin belongs to the rhythmic condition. Specifically, 4 bins in the rhythmic condition were slower than any of the bins in the arrhythmic condition.

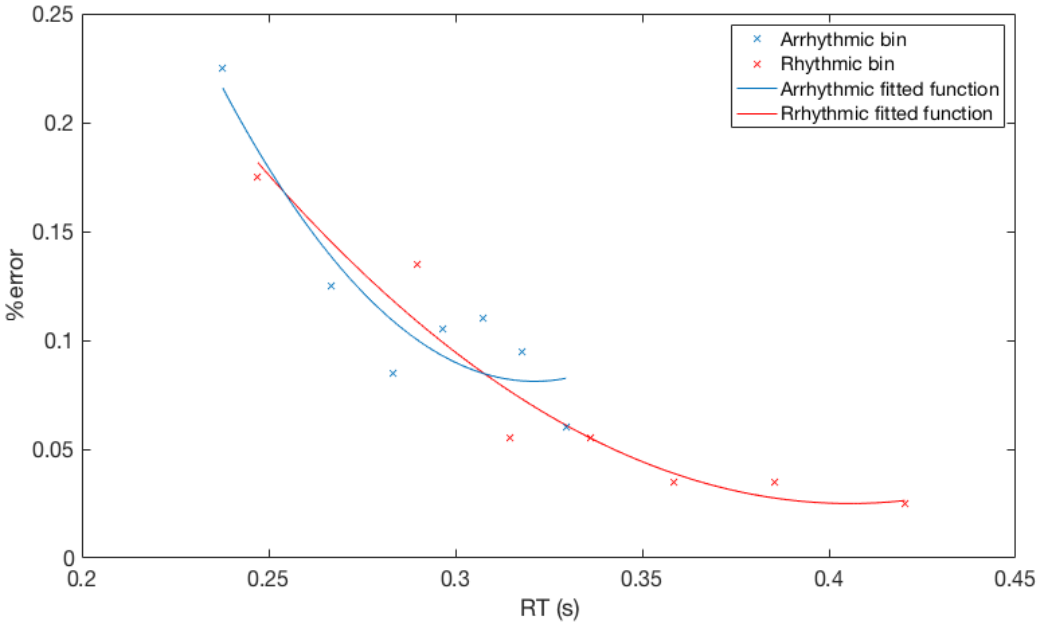


Figure 58.3. Mean RT for each bin and the corresponding error percentage for t rhythmic (red) and arrhythmic trials (blue)

Study 2: Randomized design

Response Times (RT)

Mean RT for the rhythmic condition was 0.340s (SD 0.031) and the mean RT for the arrhythmic condition was 0.337s (SD 0.032). The difference in RT between rhythmic and arrhythmic conditions was significant (p= 0.009). Figure 5.4 shows boxplots for the RT in each condition. Mean RT corroborate that participants were still receiving stimulation while they were making a response.

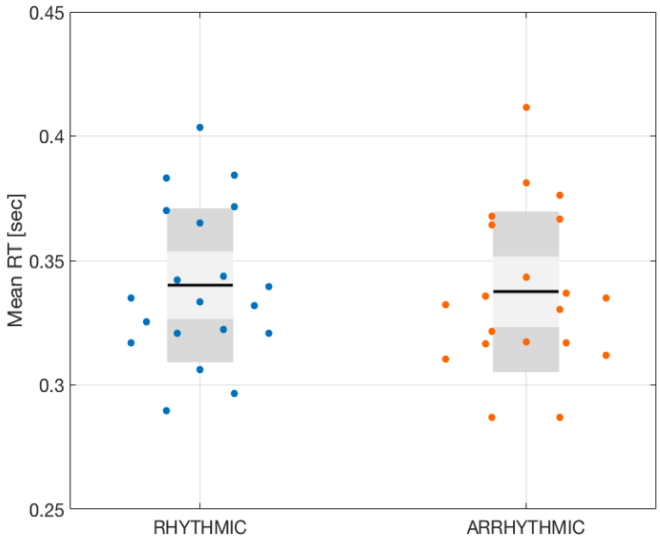


Figure 59.4. Boxplot of the RT for each condition. Every dot represents one subject's average RT

Incorrect trials

In the rhythmic condition, the mean number of incorrect trials was 11.7 (SD 7.20) and in the arrhythmic condition, the mean number of incorrect trials was 11.7 (SD 7.980).

RT including correct and incorrect trials were rank-ordered in an ascendant order and grouped in 7 bins of 20 trials for each condition. The mean RT and the percentage of incorrect trials was calculated for each bin. Figure 5.5 shows a plot with the mean RT of each bin and the corresponding error percentage for both rhythmic and arrhythmic trials. A quadratic polynomial curve has been fitted to the data.

In the rhythmic condition, the bin range goes from 0.248s to 0.413s, while in the arrhythmic condition, the bin range goes from 0.245s to 0.327s. The fastest bin belongs to the arrhythmic condition while the slowest bin belongs to the

rhythmic condition. Specifically, 4 bins in the rhythmic condition were slower than any of the bins in the arrhythmic condition.

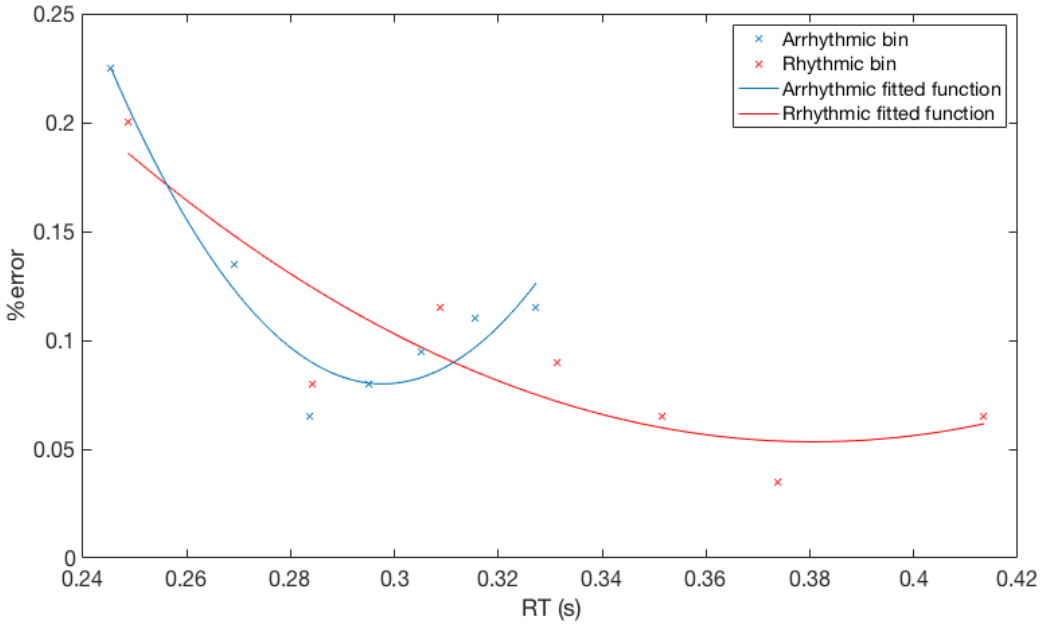


Figure 5.60. Mean RT for each bin and the corresponding error percentage for the rhythmic (red) and arrhythmic trials (blue)

5.4. Discussion

These two studies show, for the first time, that the entrainment at the mu band using MNS slows down movement initiation in healthy participants. Concretely, these two studies show that the delivery of rhythmic 12Hz MNS slows down motor response compared to arrhythmic MNS in healthy subjects.

Behavioural effects of entrainment in the motor cortex have been shown in previous studies. For example, Joundi et al. (2012) showed that entrainment of the motor cortex using tACS at the beta frequency band reduced force generation, and Pogosyan et al. (2009) showed that entrainment of the motor cortex using tACS at the beta frequency band slowed down velocity of voluntary movement. However, these studies used the conventional non-invasive brain stimulation techniques (i.e. tACS) and entrained at frequencies in the beta

band. The studies presented here show, for the first time, that (I) entrainment at the mu band in the motor cortex has a behavioural effect in healthy subjects, and (II) entrainment using MNS has a behavioural effect in healthy subjects.

Effects of mu entrainment on behaviour

Previous studies examining the effects of entrainment in the motor cortex have focused on the beta band because of its strong association with voluntary movement execution. The studies presented here prove that mu rhythm is also strongly associated with voluntary movement execution and therefore, should be given as much attention. In the two mentioned studies (Joundi et al., 2012; Pogosyan et al., 2009), stimulation at 20Hz did have an effect on the quality of the movement (i.e. velocity and force) but did not have an effect on the initiation of the movement (i.e. reaction time**)**. Results strongly support the involvement of mu-rhythm in movement preparation.

The slowing of movement initiation due to the mu synchronization is likely explained by the synchronization in the mu band reducing cortical excitability of the motor cortex and increasing the threshold of desynchronization needed to initiate the movement. This is easier to understand when it is explained what happens at a cellular level. When a population of cells fire in synchrony there is low capacity of encoding information, however, when a population of cells fire independently the capacity of encoding information within the population rises. It is likely that the mu ERD prior to movement execution is related to the increase in the capacity of information processing needed in order to execute the movement that is about to be made. The increase in synchronization produced by the rhythmic stimulation leads to a lower capacity of information processing (i.e. reduced cortical excitability) than at rest, incrementing the time needed to increase the capacity of information processing in order to initiate the movement.

Effect of MNS on behaviour

The fact that rhythmic MNS can have an effect on motor behaviour, similarly to TMS and tACS, can help improve the study of the role of brain oscillations on movement. First, MNS targets the SMC, while TMS and tACS are less specific. Second, the fact that MNS can be used, instead of TMS and tACS, allows researchers to study the role of brain oscillations on movement without the need of screening participants. For example, MNS will let us recruit people that have had a seizure in the past, people with implanted devices in their body, people that have had a head injury, and people that suffer from migraines. Also, even though TMS and tACS can be used on children, the fact that MNS stimulates the wrist instead of the brain is likely to be less concerning for parents. Third, the use of MNS will allow us to design studies with an easier and faster set up than the one used in TMS and tACS.

Limitations

Results show that rhythmic stimulation delays movement initiation, however, the delay is minimum (3ms in the randomized design and 6ms in the blocked design). Because of that, this delay would not be expected to have an impact on motor functioning. However, since no quality of the response (e.g. force) was measured, it is unknown whether the stimulation had a secondary effect on the quality of the movement.

One might argue that rhythmic stimulation had an effect on attention or visual processing instead of an effect on movement, that the arrhythmic stimulation did not have. However, there was no difference between the number of incorrect trials between rhythmic and arrhythmic stimulation. And, as expected, the fastest bins in both conditions presented the higher error percentage. Therefore, incorrect trials were likely due to a too fast response and entrainment did not affect attention or visual processing.

5.5. Conclusion and continuance

These two studies show that mu entrainment in the motor cortex delivered with MNS has a behavioural effect in motor responses by slowing them down.

The following chapter will explore the effects of rhythmic MNS on tics and urges in subjects with TS.

Note - This study was included in the published paper:

Maiquez, B. M., Sigurdsson, H. P., Dyke, K., Clarke, E., McGrath, P., Pasche, M., ... & Jackson, S. R. (2020). Entraining Movement-Related Brain Oscillations to Suppress Tics in Tourette Syndrome. *Current Biology*

CHAPTER 6

Chapter 6 - Exploring MNS clinical effects of mu entrainment

6.1. Introduction

It is known that brain oscillations play a role in cognitive functions. Specifically, mu oscillations have shown to decrease in power with increased excitability in the somatosensory cortex in order to facilitate processing of sensory information (S. Yin, Liu, & Ding, 2016). The somatosensory cortex is the area responsible for processing sensory information and is associated with premonitory urges. Therefore, by decreasing excitability in the somatosensory cortex, we would expect to suppress premonitory urges. And, since very often subjects refer to their tics as a response to their PU (Leckman et al., 1993), we would expect that the suppression of PU would lead to the suppression of tics.

Also, motor cortical excitability has been found to be positively correlated with tic severity (Orth, Münchau, & Rothwell, 2008), and a TMS study has shown that when subjects suppress their tics, they do so by decreasing motor cortical excitability (Ganos et al., 2018). Mu desynchronization has been found to be positively correlated with corticospinal excitability in the SMC (Thies et al., 2018), therefore, by decreasing excitability in the motor cortex we would expect to suppress tics.

Summary and aims

In Chapter 4, I have shown that entrainment of mu oscillations in the SMC using MNS decreases motor cortical excitability, and in Chapter 5, I have shown that entrainment of mu oscillations in the SMC using MNS has an effect on motor behaviour. In this Chapter I will deliver constant MNS to subjects with TS with

the aim to explore whether decreased cortical excitability evoked by rhythmic stimulation of the median nerve has an effect on the execution of tics in subjects with TS.

I hypothesize that rhythmic MNS will led to the reduction of tics by either reducing the urge to tic or directly reducing the tic execution.

6.2. Methods

Participants

19 participants (11 males) between 12 and 51 (mean age of 21) took part in the study. All participants had a confirmed diagnosis of TS. Participants were recruited from our clinic database and *Tourette's Action*. All participants (or a parent/guardian) signed the consent form of the study, approved by the Ethics Committee in Nottingham University and they were given 6 pounds for their time. Participants with comorbidities were not excluded. A recruitment criterion was that participants had to present very frequent tics as of every minute of the day to take part in the study.

Three of the youngest participants recruited (male, 12; male, 13; and female, 14) found the stimulation too painful and so withdrew from the study. I tested a total of 16 participants, which now included 9 males between 14 and 51 (mean age of 22). Of these 16 participants, 7 were currently taking medication, as follows: one was taking Sulpiride (antipsychotic), one was taking Clonidine (antihypertensive), one was taking Mirtazapine (antidepressant), one was taking Co-codamol (analgesic), and one was taking Citalapran (antidepressant), one was taking Fluoxetine (antidepressant) and Lorazepam (psychoactive), and one was taking Clonidine and Sertraline (antidepressant).

Their WASI scores varied from 81 to 131 with an average of 101. The total YGTSS score (100/100) varied from 21 to 100 with an average of 66 and the YGTSS score without taking into account their self-impairment measure

(50/100) varied from 21 to 50 with an average of 37. Their PUTS (32/32) score varied from 6 to 30 with an average of 20.

Results of the scores can be found in Table 6.1.

Table 6.10. Participant characteristics

TS ID	Age	Gender	Medication	Comorbidity	WASI	PUTS (max32)	Impairment	Motor	Vocal	Global
TS062	22.82	M	None	None	131	14	17	18	20	55
TS184	20.25	M	Sulpiride 400mg daily	None	82	29	50	25	25	100
TS196	37.30	F	None	None	120	22	25	25	25	75
TS061	15.00	M	None	None	99	29	0	18	16	34
TS204	32.95	M	Clonidine 50 micrograms	None	130	11	5	17	10	32
TS205	17.46	M	None	None	96	24	0	21	0	21
TS203	16.88	M	None	None	95	15	25	25	25	75
TS207	14.70	M	None	None	97	13	15	14	12	41
TS209	19.66	F	Mirtazapine 30mg	Depression, anxiety, BPD	111	30	25	19	12	56
TS198	25.13	F	Co-codamol 30mg/500mg (6x)	Depression	84	28	50	22	22	94
TS212	15.44	F	None	Ehler Danlos syndrome	110	29	49	19	13	81
TS213	21.39	F	Citalapran 20mg/day	None	109	22	45	22	16	83
TS230	20.16	F	None	None	108	18	40	22	13	75
TS231	19.36	F	None	None	84	6	50	22	14	86
TS233	13.57	M	Clonidine 50microgramsx2/day, Sertraline 25mg per day	None	81	13	35	22	21	78
TS234	51.39	M	Fluoxetine 60mg, Lorazepam 1.5mg	None	90	25	35	18	18	71
Average	22.72	-	-	-	101.69	20.50	29.13	20.56	16.38	66.06

*Greyed out area indicates missing score. WASI: Wechsler's Abbreviated Scale of Intelligence. PUTS: Premonitory Urge for Tics Scale (max score is 32). BPD: Borderline Personality Disorder. *Score taken from YGTSS: YGTSS Global Tic Severity Scale.*

Stimulation

The duration of a pulse was 200μs. The intensity of the stimulation used was the minimum intensity at which a thumb twitch was seen. However, in 3 participants, the stimulation was delivered at 80% of the intensity at which a thumb twitch was seen. This was done because these 3 participants found the stimulation too painful at the minimum intensity at which a thumb twitch was seen.

Design

The study consisted of three different stimulation conditions. In the rhythmic condition, pulses were delivered at a frequency of 10Hz (every 100ms); in the arrhythmic condition, the same number of pulses as the rhythmic condition were delivered but in random intervals of time; in the baseline condition, no stimulation was delivered. Every condition lasted 1 minute, this supposed delivering a total of 600 pulses in both the rhythmic and arrhythmic conditions. Every block included each of the three conditions repeated twice in a randomized way. There was a total of 3 blocks. After every block, participants were able to take a break, if needed. Figure 6.1 shows an example of a scheme of the design of the study for one block.

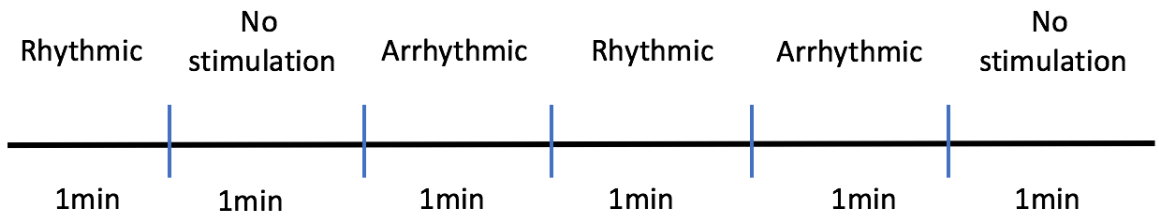


Figure 63.1. Scheme of the design of the study

Participants were asked to rate the strength of their urges using an electronic slider placed in front of them using their left hand. The further left of the slider supposed no feeling of premonitory urges at all and the further right of the slider

supposed the maximum strength, referred as unbearable, of premonitory urges that could be felt by the participant. Videos of the participants' faces as well as their upper bodies were recorded during the whole study using a web camera (Logitech C920 PRO HD Webcam) placed in front of them.

Devices

The stimulator and the slider were controlled by a bespoke MATLAB vR2017a script on a MacBook laptop running Windows 8.

Experimental procedure

Participants were seated in a small room, with their right hand resting comfortably on a blanket.

The starting intensity threshold was determined with a trial and error procedure with one pulse until a noticeable thumb twitch was seen. This threshold was further validated with a train of 8 pulses at 12 Hz frequency. Intensity was adjusted if needed.

The researcher left the room for the duration of each block in order to not have any effect on the participant's tics. The parent was allowed to stay in the room, when that happened, both the participant and the parent were asked to remain silent during the testing.

At the end of the experiment, participants were asked the following questions:

- Did you notice anything about the stimulation?
- Was the use of the slider a good way of measuring your urges/an accurate measure?

Data analysis

Video data

Both motor and vocal tics were counted and the timings of their appearance in the video were noted on an excel sheet. Moreover, every tic was scored based in its intensity as rated in the YGTSS questionnaire. Minimal tics, were tics that are usually not noticed, scored 1; mild tics, were tics that are usually not noticed but are more forceful than minimal tics, scored 2; moderate tics, were tics that are not outside the range of normal expression, scored 3; marked tics, were tics that are of an exaggerated character, scored 4; and severe tics, were tics that caused injury, scored 5.

All this was done by the researcher, who was blind to the order of conditions and therefore she didn't know whether the participant was receiving stimulation or not. The time lag between the start of the video recording and the delivery of the pulses was always removed.

Slider data

The slider output was measured in voltage, recording 20 scans per second. The percentage of voltage was calculated for each of the 18 minutes (the total voltage for each minute was added and divided by the maximum voltage for one minute and multiplied by 100 to calculate the percentage) and the average of the percentage for each condition was then calculated.

6.3. Results

After observing the video data, it was noticeable that at the beginning of every stimulation condition (either rhythmic or arrhythmic) participants took some time in order to (1) react to the stimulation, (2) think about any changes in their urges, and (3) decide to move the slider accordingly. This could take participants up to 20 seconds. For this reason, only the lasts 40 seconds of every condition were analysed in the overall data.

Tic count

The average number of tics between participants was 87 in the rhythmic condition, 87 in the arrhythmic condition and 126 in the no stimulation condition. The difference in the number of tics was significantly different between conditions (F=4.082, p=0.027), where the rhythmic condition showed less amount of tics than the no stimulation condition (t(15)=-1.992, p=0.032) and the arrhythmic condition showed less amount of tics than the no stimulation condition (t(15)=-2.145, p=0.024). There was no difference in the number of tics between the rhythmic and arrhythmic conditions (t(15)=-0.056, p=0.955). Figure 6.2 shows the number of tics for each participant in each condition.

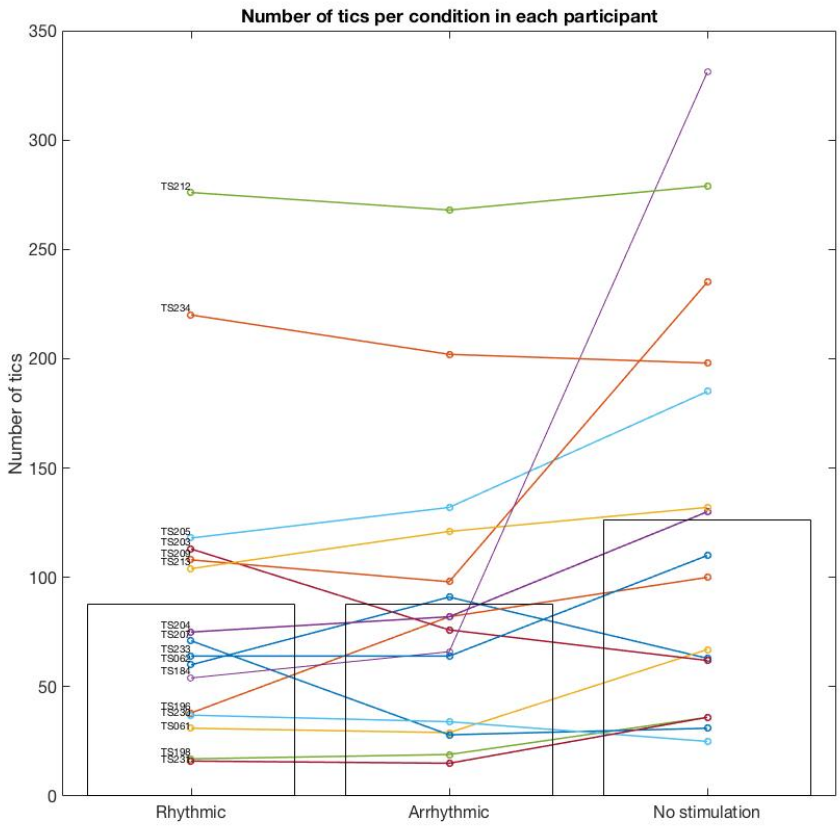


Figure 62.2. Average number of tics in each condition. The plot also shows the number of tics in each participant at each condition

Overall, the rhythmic condition supposed a reduction of 30.59% in the number of tics and the arrhythmic condition supposed a reduction of 30.34% in the number of tics.

Tic intensity

The average intensity of tics between participants was 2.76 in the rhythmic condition, 2.66 in the arrhythmic condition and 2.96 in the no stimulation condition. The difference in the intensity of tics was significantly different between conditions (F=3.905, p=0.031), where the rhythmic condition showed less amount of tics than the no stimulation condition (t(15)=-2.049, p=0.029) and the arrhythmic condition showed less amount of tics than the no stimulation condition (t(15)=-2.535, p=0.011). There was no difference in the number of tics between the rhythmic and arrhythmic conditions (t(15)=0.905, p=0.379). Figure 6.3 shows the intensity of tics for each participant in each condition.

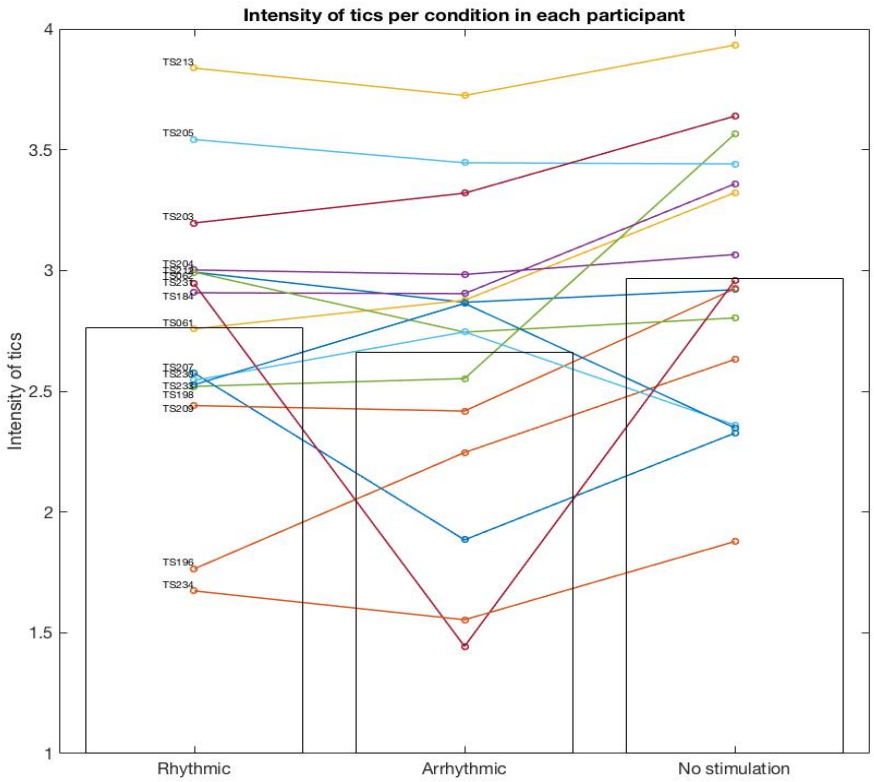


Figure 63.3. Average intensity of tics in each condition. The plot also shows the number of tics in each participant at each condition

Overall, the rhythmic condition supposed a reduction of 6.85% in the intensity of tics and the arrhythmic condition supposed a reduction of 10.31% in the intensity of tics.

Ratings of intensity of the urge to tic

The slider data from 2 participants was lost due to technical issues and so the slider data was analysed using 14 subjects. The average of the ratings in intensity of the urge to tic between participants was 27.02 in the rhythmic condition, 28.34 in the arrhythmic condition and 39.98 in the no stimulation condition. The difference in the urge to tic ratings was not significantly different between conditions (F=1.722, p=0.198). Figure 6.4 shows the intensity of urges for each participant in each condition.

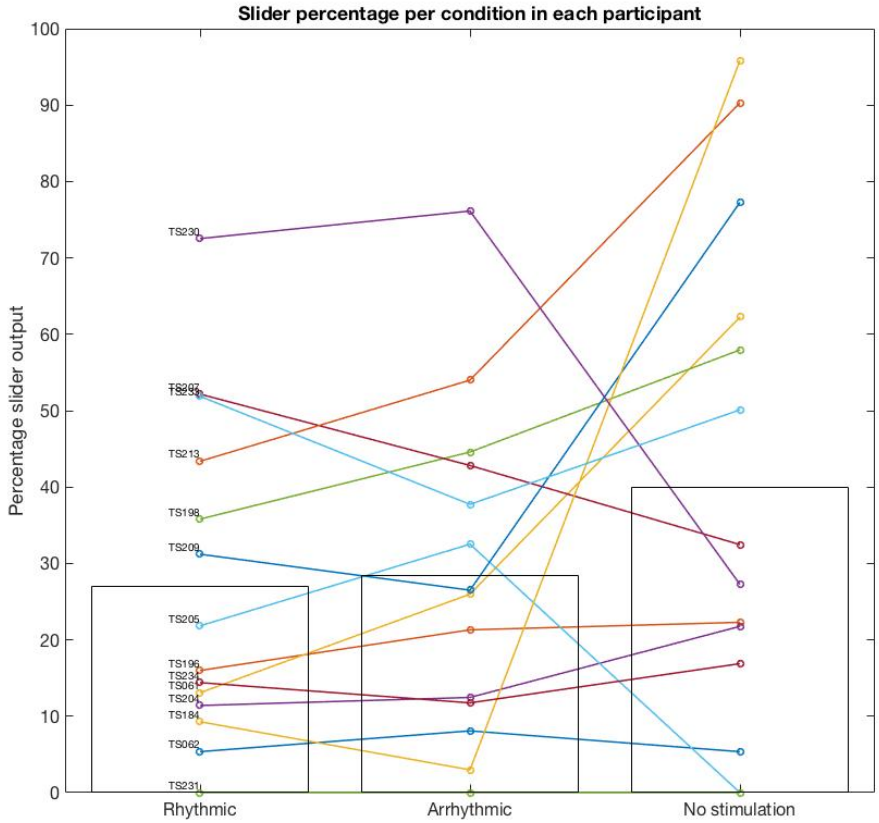


Figure 64.4. Average of the ratings in intensity of the urge to tic in each condition. The plot also shows the number of tics in each participant at each condition

Overall, the rhythmic condition supposed a reduction of 32.41% in the intensity of urges and the arrhythmic condition supposed a reduction of 29.10% in the intensity of urges.

Tic reduction vs urges reduction

A Pearson's correlation revealed that tic reduction in the rhythmic condition was significantly correlated with the reduction in urges ($r=0.723$, $p=0.0034$); and tic reduction in the arrhythmic condition was significantly correlated with the reduction in urges ($r=0.769$, $p=0.0013$).

Participants' comments on the effect of the stimulation on their tics and urges

Some participants said the stimulation had an effect on their tics, some participants said the stimulation had an effect on their urges and other participants said the stimulation had an effect on both tics and urges. Participant's comments were mainly on the effects of the stimulation on their urges or their need to tic: one participant said that muscles that never relax did so on the last block when there was stimulation, three other participants said the stimulation made them calmer, another participant said the stimulation definitely decreased her urges, another participant said stimulation stopped him from wanting to tic, another participant said with the stimulation, he didn't need to tic as much, another participant said that his urges were reduced and his tics weren't on his mind. Interestingly, one participant said that the stimulation reduced a lot her urges but not her tics and so with the stimulation, 'she could not tell when tics were going to happen'. Other comments were made on the effect of the stimulation on reducing their tics: one participant said the stimulation stopped him from ticcing, another participant said with the stimulation, he didn't tic as much, another participant said the stimulation made him tic less, another participant said the stimulation had an effect on his tics.

One participant made a comment about how his big tics didn't come out as much with the stimulation.

Three participants wondered if the reduction of tics was due to the distraction of the stimulation. Three participants mentioned that the effects of the stimulation lasted for some time after it had been stopped.

Slider reliability

Participants were asked whether the use of the slider was an accurate measure to rate their urges. Of the sixteen participants that were tested, eight participants agreed that the use of the slider was an accurate measure for their urges. One participant pointed out that if the tic was on his left hand (hand used to use the slider) or if he had bouts of tics, he was not able to use the slider so that made the measure less accurate. Another participant said it is difficult to rate the urge once the urge starts. Another participant said in order to rate the urges more accurately it would be more useful to rate the urges after every minute instead of doing it constantly. Two participants said that the use of the slider to rate the urges was accurate for half of the tics, as for the other half, the urge was too fast.

There was a significant correlation between slider output at the condition with no stimulation and PUTS scores ($r=0.591$, $p=0.025$).

6.4. Discussion

MNS was delivered in subjects with TS while they were being video recorded and while they were rating the intensity of their urges using a slider. Tic count and tic intensity rating revealed that MNS has an effect on tics. Specifically, results showed that MNS reduced the number of tics and the intensity of tics. The slider output revealed a reduction in the strength of the urges, although it failed to reach significance.

There was a noticeable variability in the effects of the stimulation between subjects. This was expected since the phenotype of TS is very different among individuals and stimulation was being delivered using the same parameters (intensity and frequency) to each subject. Further work is needed to explore the variability in the effect of the stimulation between individuals.

Unexpectedly, both the rhythmic and arrhythmic stimulation had a very similar effect. This was surprising, since each type of stimulation has a different effect in the motor cortex (see Chapter 4) and a different effect on motor behaviour (see Chapter 5). This suggests that, despite that the arrhythmic stimulation is not delivered at a specific frequency, the stimulation might also be resetting the phase of brain oscillations in the SMC, and thus decreasing cortical excitability for a short period of time, long enough to have an effect on tic execution.

The significant correlation between the reduction of urges and the reduction of tics suggest that the stimulation is having an effect on the urge to tic by suppressing it, resulting in the suppression of tics. The fact that the intensity of tics was reduced supports this, since it is known that 'bigger' tics (e.g. tics involving the head, neck, shoulders or midline abdomen) are most frequently preceded by urges than simple tics (e.g. tics involving eye blinking or mouth movements) (Cavanna et al., 2017). Participant's comments also seem to suggest that the stimulation was mainly having an effect on the urges and the reduction of tics was just a secondary effect. This was highlighted by comments like "With the stimulation, I didn't need to tic as much" or "The stimulation stopped me from wanting to tic". This is in line with the fact that most subjects with TS report their tics being a response to their urges (Leckman et al., 1993). Interestingly, there was a participant that reported that the stimulation made her urges disappear, but not her tics. Further research is needed to examine the relationship between urges and tics, and further research is needed to explore the effects of the stimulation on the experience of the urge to tic.

Three young participants found the stimulation excessively painful. This was discouraging since the subjects with most needed treatment are young children.

Further work is needed in order to explore whether the high intensity of the stimulation delivered in this study is in fact needed in order for the electric pulses to reach the SMC and thus produce similar clinical effects.

Some participants wondered whether the effect of the stimulation was due to a distraction of the sensory stimulus. Since both rhythmic and arrhythmic stimulations produced a similar effect, it could be that the stimulation was just distracting subjects from ticcing or from the urge to tic. However, some participants mentioned that the effects of the stimulation were noticed even after the stimulation had been stopped, suggesting that the effects of the stimulation were not due to a distraction.

Limitations

The fact that some participants reported that the reduction in tics and urges happened when the stimulation was on but also when the stimulation was off leads to think that the stimulation evoked aftereffects in some participants. This could have affected the results by showing less difference in the number of tics, the intensity of tics and the intensity of their urges between stimulation and no stimulation conditions. Further research is needed to explore the after-effects of the stimulation.

Most participants agreed that the use of the slider was a good measure of the intensity of the urge to tic. This is supported by the significant correlation between slider output during no stimulation and PUTS scores. However, it is probably that some participants used the slider in the opposite way. This is likely for participant TS205, whose tics were reduced with the stimulation, but they reported higher urges than when there was no stimulation delivered. This and the fact that most of the participants pointed out a reduction in urges evoked by the stimulation but there was no significant difference in the urge intensity between stimulation and no stimulation conditions, makes the slider results unreliable and so they should be taken with caution.

6.5. Conclusion

MNS significantly reduces the number of tics and the intensity of tics in subjects with TS. A correlation between the urge to tic and tic frequency suggest that the reduction of tics is achieved by the electric stimulation having an effect on the urge to tic by suppressing it, resulting in the suppression of tics. These findings suggest that MNS has the potential to be used as a safe and effective treatment for TS.

Note - This study was included in the published paper:

Maiquez, B. M., Sigurdsson, H. P., Dyke, K., Clarke, E., McGrath, P., Pasche, M., ... & Jackson, S. R. (2020). Entraining Movement-Related Brain Oscillations to Suppress Tics in Tourette Syndrome. *Current Biology*

CHAPTER 7

Chapter 7 - General Discussion and Thesis conclusion

Within this thesis I have used neuroimaging techniques to explore brain activity preceding voluntary movement and tics in people with Tourette syndrome and used non-invasive brain stimulation to modulate this brain activity. The results of the studies presented in this thesis have supported previous findings as well as provided new knowledge on brain activity preceding tics and voluntary movement in subjects with TS; and have shown the effects in the brain of a technique based on peripheral nerve stimulation, as well as the effects that it generates on the execution of voluntary movements and the execution of tics.

I will summarize each of the main findings in this discussion. I will first discuss the findings on brain activity preceding tics and voluntary movement and how these relate to each other; and then I will discuss the modulation of brain activity using MNS as a treatment for TS.

7.1. Activity in the brain before tics and voluntary movement

BOLD activity as well as ERP preceding tics has been explored in many different studies. Only one study has explored cortical activity preceding tics using EEG/MEG (Niccolai et al., 2019), but results are unreliable due to the fact that the analysed tics occurred while performing a computer motor task and therefore data is likely to be contaminated by the effects of movement preparation and post-movement beta rebounds. The fact that cortical excitability plays an important role in tic execution, motivated us to explore cortical activity prior to tics by means of the changes in spectral power of brain oscillations. In Chapter 3a.2, I recorded EEG while subjects with TS were at rest. They were asked not to do any effort to suppress their tics and not to perform any voluntary movements. Results showed no consistent pattern of

cortical activity preceding tics suggesting that different tics are preceded by different patterns of cortical activity. Although all tics are spontaneous involuntary movements, their heterogeneity (i.e. they can be preceded by PU or not; they can be simple motor tics, simple vocal tics, complex motor tics or complex vocal tics) is likely reflected in the high variability of the pattern of cortical activity preceding tics. Despite that there was no consistent pattern of cortical activity preceding tics, it was common to see an increase in excitability within one second before the tic onset in single tic analyses.

The execution of voluntary movements can suppress tics. This motivated us to explore cortical activity preceding voluntary movements in subjects with TS using EEG. In Chapter 3a.1, I explored motor cortical activity preceding voluntary finger movements in subjects with TS and HS. Even though voluntary movement between HS and subjects with TS was not directly compared, results showed that voluntary movements are characterized by a decrease in mu power in HS but not in subjects with TS, suggesting that mu rhythm is involved in the characterized reduced excitability preceding and during voluntary movement in subjects with TS found in previous MRI and TMS studies (e.g. Heise et al., 2010; Thomalla et al., 2014). These results support previous suggestions (Draper et al., 2015) that subjects with TS need to increase their cortical inhibition in order to generate the desired motor response. Further analyses showed a positive correlation between mu power and tic severity, suggesting that subjects with higher tic severity require a higher reduction in excitability than subjects with lower tic severity, in order to perform the desired voluntary movement.

How different tics are from voluntary movements is a question that has been in the literature for a long time. The ERP and time-frequency results from Chapter 3a.1, together with the ERP and time-frequency results from Chapter 3a.2 showed clear evidence that voluntary movements and tics are preceded by different brain activity in the motor cortex. The evidence was characterised by the presence of BP preceding voluntary movement but not tics, and the presence of beta ERD preceding voluntary movement but not tics.

In order to examine the areas being activated preceding tics, in Chapter 3b fMRI was used while participants were at rest. In this case participants were asked to suppress their tics to avoid big movements inside the scanner. PU precede tics and are very often referred to as the cause of tics, therefore the aim of the study was to explore the brain activity at the individual's subjective strongest PU. This was done by asking participants to press a button whenever they felt a strong urge to tic. The brain activity related to the button press was later subtracted in the analyses. The activity preceding tic execution has previously been examined in imaging studies (Bohlhalter et al., 2006; Neuner et al., 2014), however, there are no studies investigating the state of the SMC at the strongest PU. Disappointingly, results of this study did not show brain activity at the strongest PU. It is likely that either the baseline was contaminated by PU or participants were not feeling strong enough urges due to the short time period between their tics. Since subjects were not very successful in suppressing their tics, the BOLD activity 1 and 2 seconds before tics was analysed. Results showed that tics are preceded by an initial activity in the right primary somatosensory cortex 2 seconds before tic onset, and right primary motor cortex 1 second before tic onset.

All these findings suggest that, as reported in previous studies, increased cortical excitability in the SMC is related to the execution of tics and decreased cortical excitability in the SMC is related to the suppression of tics.

7.2. Modulation of the brain activity in the motor cortex as a treatment for TS

It is thought that the execution of tics is the consequence of the abnormal spontaneous activity in the striatum leading to an overactivity in cortical motor areas. In fact, it has been shown that the suppression of tics is achieved by reducing cortical excitability (Ganos et al., 2018). Given that increase in phase synchrony of brain oscillations results in a decrease in cortical excitability,

entrainment of brain oscillations decreases cortical excitability. This has been shown by delivering rhythmic TMS pulses on the parietal cortex (Thut et. al, 2011).

Median nerve stimulation has an effect on the SMC (Truett Allison et al., 1992). The fact that MNS is a non-invasive brain stimulation technique that, contrary to TMS and tDCS, does not require screening and can be applied outside the clinic, motivated us to explore whether MNS could be used to entrain brain oscillations in the SMC. To date, no study had considered to explore this. In Chapter 4, I delivered rhythmic MNS at two different frequencies belonging to two different frequency bands (i.e. mu and beta) while recording EEG in HS and showed evidence that rhythmic MNS did entrain brain oscillations in the motor cortex. This evidence was characterized by an increase in EEG power and ITC at the frequency of stimulation that happened during rhythmic MNS but not during arrhythmic MNS.

It is known that brain oscillations play a role in cognitive functions and that the entrainment of brain oscillations has an effect on behaviour. This motivated us to explore the effects of entrainment using MNS on behaviour. In Chapter 5, I delivered rhythmic MNS at a frequency in the mu band while HS were performing a motor task on the computer and showed evidence that rhythmic MNS slowed down motor response. This evidence was characterized by a decrease in response time of 3-6ms when rhythmic MNS was delivered compared to when arrhythmic MNS was delivered. This study not only showed that entrainment using MNS has a behavioural effect, but it also showed the importance of mu rhythm in movement initiation.

In Chapter 6, I delivered constant rhythmic MNS to subjects with TS and monitored their tics with a video camera and asked them to rate the strength of their urges using an electronic slider. Results showed that MNS reduced the number of tic execution and the intensity of tics; and a correlation between the urge to tic and tic frequency suggested that the reduction of tics is achieved by the electric stimulation having an effect on the urge to tic by suppressing it, resulting in the suppression of tics.

7.3. Thesis limitations and Future work

TS is a very complex disorder. Individuals with TS can have a very different phenotype. For example, they can present comorbidities including ADHD, OCD, ASD, and impulsivity, between others; they can present or not urges; they can have a huge variety of tics involving simple motor and vocal tics and complex motor and vocal tics. Also, tics can be characterized very differently within participants. For example, some tics can be preceded by urges while others are not, and when the urge is present it can be either strong or weak in intensity. I have recruited participants with TS that presented comorbidities as well as participants with TS that were taking medication at the time of the testing. Although this limits the study of TS on its own, this sample is representative of the TS population, and the execution of tics should be preceded by similar brain activity and tic suppression should be performed similarly by suppressing motor output. On the other side, I have also recruited participants with TS that either presented or lacked urges and had a huge range of tics varying in complexity and intensity, which could be preceded or not by urges. This variability could have altered the results, specifically in the study of brain activity preceding tics in Chapter 3a.2 and Chapter 3b. I would suggest that future research focus on a specific type of tic with similar properties instead of including all general tics in the same analyses. Furthermore, some individuals report to feel a relief of the urge after the tic is executed, while other individuals report to feel the urge to tic disappear temporally after tic execution. In Chapter 3b no significant BOLD activity was found at the individual's subjective strongest PU. I would suggest that future research investigating activity associated with the PU using fMRI should only recruit participants whose PU temporally disappear after the execution of the tic in order to have a similar subgroup and a baseline temporally free of urges.

Results in Chapter 3a.1 have shown us that it is important to have a heterogeneous group when it comes to severity of the disease as the recruitment of only subjects with low severe TS or only subjects with high severe TS can mislead the final results. For example, recruiting a group of

subjects with low severe TS would likely give us results showing similar brain activation between HS and subjects with TS during voluntary movement (both groups showing a decrease in both mu and beta power), while recruiting a group of subjects with high severe TS would likely give us results showing different brain activation between HS and subjects with TS during voluntary movement (HS group showing a decrease in both mu and beta power and TS group showing a decrease in beta power but not in mu power). I would suggest that future research with TS recruit participants with either mixed severity of the disease in order to have a representative sample of the TS population, or consider low severe TS and high severe TS as two different subgroups within the TS group.

In Chapter 4 I have shown how it is possible to use MNS to entrain the SMC and in Chapter 5 I have shown how it is possible to use MNS to explore the effects of brain oscillations on behaviour. Future research exploring the effects of entrainment on cognitive functions would benefit from using this technique as it decreases set up time and increases potential subjects that can be tested compared to TMS and tDCS/tACS. However, it is important to point out that MNS limits to the study of brain oscillations in the SMC, while TMS and tDCS can be used in other brain regions.

The research in Chapter 6 suggests that the execution of most tics is the result of the feeling of the urge to tic, since the reduction of urges is correlated with a reduction in tics. Some tics are preceded by urges while other tics are not. Interestingly, it is not always the case that getting rid of the urge will lead to no execution of tics in an individual, as one participant reported in Chapter 6. This suggest that the feeling of urge to tic and tic execution is not always linked. Further research is needed to explore the relationship between urges and tics in TS.

7.4. Conclusion

In this thesis I used neuroimaging techniques to explore brain activity preceding voluntary movement and tics in people with Tourette syndrome and used non-invasive brain stimulation to modulate this brain activity and generate an effect on the execution of voluntary movements and the execution of tics.

The research revealed that the activity in the brain before tics is located in motor and sensory areas, and it varies between tics, likely due to the heterogeneity of tics. The activity in the brain before voluntary movements, contrary to HS, does not show an increase in excitability in the mu band in the SMC, and this varies between subjects in that subjects with a more severe disease show an increase in inhibition while subjects with a less severe disease show an increase in excitability. All these findings suggest that, as reported in previous studies, increased cortical excitability in the SMC is related to the execution of tics and decreased cortical excitability in the SMC is related to the suppression of tics.

Following this research, I showed that we are able to modulate cortical activity using MNS by delivering rhythmic electric stimulation at the wrist. Using EEG, I showed that this modulation leads to a decrease in excitability in the SMC at the frequency of stimulation. Finally, I showed that this modulation has a behavioural effect by reducing the speed of motor response and a clinical effect by reducing the number of tics and the intensity of tics in subjects with TS. These findings suggest that MNS has the potential to be used as a safe and effective treatment for TS.

References

Allison, T, McCarthy, G., Wood, C. C., Darcey, T. M., Spencer, D. D., Williamson, P. D., & Spencer, D. D. (1989). Human cortical potentials evoked by stimulation of the median nerve. I. Cytoarchitectonic areas generating short-latency activity. *Journal of Neurophysiology*, *62*(3), 711–722.

Allison, Truett, Mccarthy, G., & Wood, C. C. (1992). The relationship between human long-latency somatosensory evoked potentials recorded from the cortical surface and from the scalp. *Electroencephalography and Clinical Neurophysiology*, *84*(4), 301–314. https://doi.org/doi:10.1016/0168-5597(92)90082-M

Allison, Truett, McCarthy, G., Wood, C. C., & Jones, S. J. (1991). Potentials Evoked in Human and Monkey Cerebral Cortex By Stimulation of the Median Nerve. *Brain*, *114*(6), 2465–2503. https://doi.org/10.1093/brain/114.6.2465

Amunts, K., & Zilles, K. (2012). Architecture and organizational principles of Broca's region. *Trends in Cognitive Sciences*, *16*(8), 418–426. https://doi.org/10.1016/j.tics.2012.06.005

Artoni, F., Menicucci, D., Delorme, A., Makeig, S., & Micera, S. (2014). RELICA: a method for estimating the reliability of independent components. *Neuroimage*, *103*, 391-400.

Balzamo, E., Marquis, P., Chauvel, P., & Régis, J. (2004). Short-latency components of evoked potentials to median nerve stimulation recorded by intracerebral electrodes in the human pre- and postcentral areas. *Clinical Neurophysiology*, *115*(7), 1616–1623. https://doi.org/10.1016/j.clinph.2004.02.012

Banaschewski, T., & Rothenberger, A. (2003). Premonitory sensory phenomena and suppressibility of tics in Tourette syndrome : developmental aspects in children and adolescents. *Development and Psychopathology*, (1994), 700–703. https://doi.org/10.1017/S0012162203001294

Baron-Cohen, S., Wheelwright, S., Skinner, R., Martin, J., & Clubley, E. (2001). The Autism-Spectrum Quotient (AQ): Evidence from Asperger Syndrome/High-Functioning Autism, Malesand Females, Scientists and Mathematicians. *Journal of Autism and Developmental Disorders*, *31*(1), 5–17. https://doi.org/10.1023/A:1005653411471

Baumung, L., Kirsten, M., Dyke, K., Jackson, G., Jackson, S., Golm, D., … Brandt, V. (2020). Developing the Premonitory Urges for Tic Disorders Scale – Revised (PUTS-R), 1–14. https://doi.org/10.1111/jnp.12216

Benjamini, Y., & Hochberg, Y. (1995). Controlling the False Discovery Rate: A Practical and Powerful Approach to Multiple Testing. *Journal of the Royal Statistical Society. Series B (Methodological)*, *57*(1), 289–300. Retrieved from http://www.jstor.org/stable/2346101

BERARDELLI, A., ROTHWELL, J. C., DAY, B. L., & MARSDEN, C. D. (1985). PATHOPHYSIOLOGY OF BLEPHAROSPASM AND OROMANDIBULAR DYSTONIA. *Brain*, *108*(3), 593–608. https://doi.org/10.1093/brain/108.3.593

Bohlhalter, S., Goldfine, A., Matteson, S., Garraux, G., Hanakawa, T., Kansaku, K., … Hallett, M. (2006). Neural correlates of tic generation in Tourette syndrome: An event-related functional MRI study. *Brain*, *129*(8), 2029–2037. https://doi.org/10.1093/brain/awl050

Brandt, Valerie C., Beck, C., Sajin, V., Baaske, M. K., Bäumer, T., Beste, C., … Münchau, A. (2016). Temporal relationship between premonitory urges and tics in Gilles de la Tourette syndrome. *Cortex*, *77*, 24–37. https://doi.org/10.1016/j.cortex.2016.01.008

Brandt, Valerie Cathérine, Beck, C., Sajin, V., Anders, S., & Münchau, A. (2016). Convergent validity of the PUTS. *Frontiers in Psychiatry*, *7*(APR), 1–7. https://doi.org/10.3389/fpsyt.2016.00051

Brittain, J. S., Probert-Smith, P., Aziz, T. Z., & Brown, P. (2013). Tremor suppression by rhythmic transcranial current stimulation. *Current Biology*, *23*(5), 436–440. https://doi.org/10.1016/j.cub.2013.01.068

Brookes, M. J., Hall, E. L., Robson, S. E., Price, D., Palaniyappan, L., Liddle, E. B., … Morris, P. G. (2015). Complexity measures in magnetoencephalography: Measuring "disorder" in schizophrenia. *PLoS ONE*, *10*(4), 1–23. https://doi.org/10.1371/journal.pone.0120991

Bullmore, E., & Sporns, O. (2009). Complex brain networks: graph theoretical analysis of structural and functional systems. *Nature Publishing Group*, *10*(3), 186–198. https://doi.org/10.1038/nrn2575

Cavanna, A. E., Black, K. J., Hallett, M., & Voon, V. (2017). Neurobiology of the Premonitory Urge in Tourette's Syndrome: Pathophysiology and Treatment Implications. *The Journal of Neuropsychiatry and Clinical Neurosciences*, appi.neuropsych. https://doi.org/10.1176/appi.neuropsych.16070141

Cavanna, A. E., & Rickards, H. (2013). The psychopathological spectrum of Gilles de la Tourette syndrome. *Neuroscience and Biobehavioral Reviews*, *37*(6), 1008–1015. https://doi.org/10.1016/j.neubiorev.2012.10.011

Chen, R., Corwell, B., & Hallett, M. (1999). Modulation of motor cortex excitability by median nerve and digit stimulation. *Exp. Brain Res.*, *129*(1), 77. https://doi.org/10.1007/s002210050938

Chen, R., Yaseen, Z., Cohen, L. G., & Hallett, M. (1998). Time course of corticospinal excitability in reaction time and self-paced movements. *Annals of Neurology*, *44*(3), 317–325. https://doi.org/10.1002/ana.410440306

Della Penna, S., Torquati, K., Pizzella, V., Babiloni, C., Franciotti, R., Rossini, P. M., & Romani, G. L. (2004). Temporal dynamics of alpha and beta rhythms in human SI and SII after galvanic median nerve stimulation. A MEG study. *NeuroImage*, *22*(4), 1438–1446. https://doi.org/10.1016/j.neuroimage.2004.03.045

Delorme, A., Fernsler, T., Serby, H., & Makeig, S. (2006). EEGLAB Tutorial. *University of San Diego California*.

Desmurget, M., & Sirigu, A. (2009). A parietal-premotor network for movement intention and motor awareness. *Trends in Cognitive Sciences*, *13*(10), 411–419. https://doi.org/10.1016/j.tics.2009.08.001

Draper, A., Jackson, G., Morgan, P. S., & Jackson, S. R. (2016). Premonitory urges are associated with decreased grey matter thickness within the insula and sensorimotor cortex in young people with Tourette syndrome. *Journal of Neuropsychology*, *10*(1), 143–153. https://doi.org/10.1111/jnp.12089

Draper, A., Jude, L., Jackson, G., & Jackson, S. R. (2015). Motor excitability during movement preparation in Tourette syndrome. *Journal of Neuropsychology*, *9*(1), 33–44.

https://doi.org/10.1111/jnp.12033

Draper, A., Stephenson, M. C., Jackson, G., Ramani, A., Morgan, P. S., Morris, P. G., & Jackson, S. R. (2014). Increased GABA contributes to enhanced control over motor excitability in tourette syndrome. *Current Biology*, *24*(19), 2343–2347. https://doi.org/10.1016/j.cub.2014.08.038

Duggal, H. S., & Nizamie, S. H. (2002). Bereitschaftspotential in Tic Disorders : A Preliminary Observation, (December), 487–489.

Enatsu, R., Nagamine, T., Matsubayashi, J., Maezawa, H., Kikuchi, T., Fukuyama, H., … Hashimoto, N. (2014). The modulation of rolandic oscillation induced by digital nerve stimulation and self-paced movement of the finger: A MEG study. *Journal of the Neurological Sciences*, *337*(1–2), 201–211. https://doi.org/10.1016/j.jns.2013.12.011

Franzkowiak, S., Pollok, B., Biermann-Ruben, K., Südmeyer, M., Paszek, J., Jonas, M., … Schnitzler, A. (2010). Altered pattern of motor cortical activation-inhibition during voluntary movements in Tourette syndrome. *Movement Disorders*, *25*(12), 1960–1966. https://doi.org/10.1002/mds.23186

Fukuda, M., Nishida, M., Juhász, C., Muzik, O., Sood, S., Chugani, H. T., & Asano, E. (2008). Short-latency median-nerve somatosensory-evoked potentials and induced gamma-oscillations in humans. *Brain*, *131*(7), 1793–1805. https://doi.org/10.1093/brain/awn100

Gaetz, W., Edgar, J. C., Wang, D. J., & Roberts, T. P. L. (2011). Relating MEG measured motor cortical oscillations to resting ??-Aminobutyric acid (GABA) concentration. *NeuroImage*, *55*(2), 616–621. https://doi.org/10.1016/j.neuroimage.2010.12.077

Ganos, C., Kahl, U., Brandt, V., Schunke, O., Bäumer, T., Thomalla, G., … Kühn, S. (2014). The neural correlates of tic inhibition in Gilles de la Tourette syndrome. *Neuropsychologia*, *65*, 297–301. https://doi.org/10.1016/j.neuropsychologia.2014.08.007

Ganos, C., Rocchi, L., Latorre, A., Hockey, L., Palmer, C., Joyce, E. M., … Rothwell, J. (2018). Motor cortical excitability during voluntary inhibition of involuntary tic movements. *Movement Disorders*, (November). https://doi.org/10.1002/mds.27479

Goodman, R. (1997). The strengths and difficulties questionnaire: A research note. *Journal of Child Psychology and Psychiatry and Allied Disciplines*, *38*(5), 581–586. https://doi.org/10.1111/j.1469-7610.1997.tb01545.x

Hashemiyoon, R., Kuhn, J., & Visser-Vandewalle, V. (2016). Putting the Pieces Together in Gilles de la Tourette Syndrome: Exploring the Link Between Clinical Observations and the Biological Basis of Dysfunction. *Brain Topography*, 1–27. https://doi.org/10.1007/s10548-016-0525-z

Heise, K. F., Steven, B., Liuzzi, G., Thomalla, G., Jonas, M., Müller-Vahl, K., … Hummel, F. C. (2010). Altered modulation of intracortical excitability during movement preparation in Gilles de la Tourette syndrome. *Brain*, *133*(2), 580–590. https://doi.org/10.1093/brain/awp299

Helfrich, R. F., Schneider, T. R., Rach, S., Trautmann-Lengsfeld, S. A., Engel, A. K., & Herrmann, C. S. (2014). Entrainment of brain oscillations by transcranial alternating current stimulation. *Current Biology*, *24*(3), 333–339. https://doi.org/10.1016/j.cub.2013.12.041

Hirata, M., Kato, A., Taniguchi, M., Ninomiya, H., Cheyne, D., Robinson, S. E., … Yoshimine, T. (2002). Frequency-dependent spatial distribution of human somatosensory evoked neuromagnetic fields. *Neuroscience Letters*, *318*(2), 73–76. https://doi.org/10.1016/S0304-3940(01)02483-1

Houdayer, E., Labyt, E., Cassim, F., Bourriez, J. L., & Derambure, P. (2006). Relationship between event-related beta synchronization and afferent inputs: Analysis of finger movement and peripheral nerve stimulations. *Clinical Neurophysiology*, *117*(3), 628–636. https://doi.org/10.1016/j.clinph.2005.12.001

Jackson, G., Draper, A., Dyke, K., Ramani, A., & Jackson, S. R. (2015). Inhibition, Disinhibition, and the Control of Action in Tourette Syndrome. *Trends in Cognitive Sciences*, *19*(11), 655–665. https://doi.org/10.1016/j.tics.2015.08.006

Jackson, S. R. (2011). On the functional anatomy of the urge-for-action. *Journal of Toxicology - Clinical Toxicology*, *39*(2), 135–142. https://doi.org/10.1081/CLT-100103829

Jackson, S. R., Parkinson, A., Jung, J., Ryan, S. E., Morgan, P. S., Hollis, C., & Jackson, G. (2011). Compensatory neural reorganization in Tourette syndrome. *Current Biology*, *21*(7), 580–585. https://doi.org/10.1016/j.cub.2011.02.047

Jackson, S. R., Parkinson, A., Manfredi, V., Millon, G., Hollis, C., & Jackson, G. M. (2013). Motor excitability is reduced prior to voluntary movements in children and adolescents with Tourette syndrome. *Journal of Neuropsychology*, *7*(1), 29–44. https://doi.org/10.1111/j.1748-6653.2012.02033.x

Joundi, R. A., Jenkinson, N., Brittain, J.-S., Aziz, T. Z., & Brown, P. (2012). Driving Oscillatory Activity in the Human Cortex Enhances Motor Performance. *Current Biology*, *22*(5), 403–407. https://doi.org/10.1016/j.cub.2012.01.024

Julie Onton & Scott Makeig, SCCN/INC/UCSD, March 2006

Jung, J., Jackson, S. R., Parkinson, A., & Jackson, G. (2012). Cognitive control over motor output in Tourette syndrome. *Neuroscience and Biobehavioral Reviews*, *37*(6), 1016–1025. https://doi.org/10.1016/j.neubiorev.2012.08.009

Karp, B. I., Porter, S., Toro, C., & Hallett, M. (1996). Simple motor tics may be preceded by a premotor potential. *J.Neurol.Neurosurg.Psychiatry*, *61*(0022-3050 (Print)), 103–106. https://doi.org/10.1136/jnnp.61.1.103

Kirschstein, T., & Kohling, R. (2009). What is the Source of the EEG ?, *40*(3), 146–149.

Leckman, J. F. (2002). Tourette's syndrome. *Lancet*, *360*(9345), 1577–1586. https://doi.org/10.1016/S0140-6736(02)11526-1

LECKMAN, J. F., RIDDLE, M. A., HARDIN, M. T., ORT, S. I., SWARTZ, K. L., STEVENSON, J., & COHEN, D. J. (1989). The Yale Global Tic Severity Scale: Initial Testing of a Clinician-Rated Scale of Tic Severity. *Journal of the American Academy of Child & Adolescent Psychiatry*, *28*(4), 566–573. https://doi.org/https://doi.org/10.1097/00004583-198907000-00015

Leckman, J. F., Walker, D. E., & Cohen, D. J. (1993). Premonitory urges in Tourette's syndrome. *The American Journal of Psychiatry*. US: American Psychiatric Assn. https://doi.org/10.1176/ajp.150.1.98

Leocani, L., Toro, C., Manganotti, P., Zhuang, P., & Hallett, M. (1997). Event-related coherence and event-related desynchronization/synchronization in the 10 Hz and 20 Hz EEG during self- paced movements. *Electroencephalography and Clinical Neurophysiology - Evoked Potentials*, *104*(3), 199–206. https://doi.org/10.1016/S0168-5597(96)96051-7

Lerner, A, Bagic, A., Boudreau, E. A., Hanakawa, T., Pagan, F., Mari, Z., … Hallett, M. (n.d.). Neuroimaging of neuronal circuits involved in tic generation in patients with Tourette syndrome. *Neurology*, *68*(23), 1979–1987. article. https://doi.org/10.1212/01.wnl.0000264417.18604.12

Lerner, Alicja, Bagic, A., Hanakawa, T., Boudreau, E. A., Pagan, F., Mari, Z., … Hallett, M. (2009). Involvement of insula and cingulate cortices in control and suppression of natural urges. *Cerebral Cortex*, *19*(1), 218–223. https://doi.org/10.1093/cercor/bhn074

Liao, W., Yu, Y., Miao, H.-H., Feng, Y. X., Ji, G. J., & Feng, J.-H. (2016). Inter-hemispheric Intrinsic Connectivity as a Neuromarker for the Diagnosis of Boys with Tourette Syndrome. *Molecular Neurobiology*, 1–9. https://doi.org/10.1007/s12035-016-9863-9

Libraries, C. C. (2006). Tourette ' s syndrome : from behaviour to biology. *Search*, *4*(March), 1–20. https://doi.org/10.1016/S1474-

Light, G. A., Williams, L. E., Minow, F., Sprock, J., Rissling, A., Sharp, R., ... & Braff, D. L. (2010). Electroencephalography (EEG) and event-related potentials (ERPs) with human participants. *Current protocols in neuroscience*, *52*(1), 6-25.

Makoto's preprocessing pipeline. (n.d.). Retrieved April 23, 2020, from https://sccn.ucsd.edu/wiki/Makoto's_preprocessing_pipeline

McCairn, K. W., Iriki, A., & Isoda, M. (2013). Global Dysrhythmia of Cerebro-Basal Ganglia-Cerebellar Networks Underlies Motor Tics following Striatal Disinhibition. *Journal of Neuroscience*, *33*(2), 697–708. https://doi.org/10.1523/JNEUROSCI.4018-12.2013

Mota, I. A., & Lins, O. G. (2017). Bereitschaftspotential preceding spontaneous and voluntary eyelid blinks in normal individuals. *Clinical Neurophysiology*, *128*(1), 100–105. https://doi.org/10.1016/j.clinph.2016.10.010

Mullinger, K. J., Mayhew, S. D., Bagshaw, A. P., Bowtell, R., & Francis, S. T. (2014). Evidence that the negative BOLD response is neuronal in origin: A simultaneous EEG-BOLD-CBF study in humans. *NeuroImage*, *94*, 263–274. https://doi.org/10.1016/j.neuroimage.2014.02.029

Neuner, I., Werner, C. J., Arrubla, J., Stöcker, T., Ehlen, C., Wegener, H. P., … Shah, N. J. (2014). Imaging the where and when of tic generation and resting state networks in adult Tourette patients. *Frontiers in Human Neuroscience*, *8*(May), 362. https://doi.org/10.3389/fnhum.2014.00362

Neuper, C., & Pfurtscheller, G. (2001a). Event-related dynamics of cortical rhythms: Frequency-specific features and functional correlates. *International Journal of Psychophysiology*, *43*(1), 41–58. https://doi.org/10.1016/S0167-8760(01)00178-7

Neuper, C., & Pfurtscheller, G. (2001b). Evidence for distinct beta resonance frequencies in human EEG related to specific sensorimotor cortical areas. *Clinical Neurophysiology*, *112*(11), 2084–2097. https://doi.org/10.1016/S1388-2457(01)00661-7

Niccolai, V., Korczok, S., Finis, J., Jonas, M., Thomalla, G., Siebner, H. R., … Biermann-Ruben, K. (2019). A peek into premonitory urges in Tourette syndrome: Temporal evolution of neurophysiological oscillatory signatures. *Parkinsonism and Related Disorders*, (March), 0–1. https://doi.org/10.1016/j.parkreldis.2019.05.039

Niccolai, V., van Dijk, H., Franzkowiak, S., Finis, J., Südmeyer, M., Jonas, M., … Biermann-Ruben, K.

(2016). Increased beta rhythm as an indicator of inhibitory mechanisms in tourette syndrome. *Movement Disorders*, *31*(3), 384–392. https://doi.org/10.1002/mds.26454

Nikouline, V. V., Linkenkaer-Hansen, K., Wikstr??m, H., Kes??niemi, M., Antonova, E. V., Ilmoniemi, R. J., & Huttunen, J. (2000). Dynamics of mu-rhythm suppression caused by median nerve stimulation: A magnetoencephalographic study in human subjects. *Neuroscience Letters*, *294*(3), 163–166. https://doi.org/10.1016/S0304-3940(00)01562-7

Obeso, J. A., & Marsden, C. D. (1981). Simple tics in Gilles de la Tourette ' s syndrome are not prefaced by a normal premovement EEG potential. *J.Neurol.Neurosurg.Psychiatry*, (44), 735–738. https://doi.org/10.1136/jnnp.44.8.735

Orth, M. (2009). Transcranial magnetic stimulation in Gilles de la Tourette syndrome. *Journal of Psychosomatic Research*, *67*(6), 591–598. https://doi.org/10.1016/j.jpsychores.2009.07.014

Orth, M., Münchau, A., & Rothwell, J. C. (2008). Corticospinal System Excitability at Rest Is Associated with Tic Severity in Tourette Syndrome. *Biological Psychiatry*, *64*(3), 248–251. https://doi.org/10.1016/j.biopsych.2007.12.009

Peterson, B. S., Skudlarski, P., Anderson, a W., Zhang, H., Gatenby, J. C., Lacadie, C. M., … Gore, J. C. (1998). A functional magnetic resonance imaging study of tic suppression in Tourette syndrome. *Archives of General Psychiatry*, *55*(4), 326–333. https://doi.org/10.1001/archpsyc.55.4.326

Pfurtscheller, G., Woertz, M., Müller, G., Wriessnegger, S., & Pfurtscheller, K. (2002). Contrasting behavior of beta event-related synchronization and somatosensory evoked potential after median nerve stimulation during finger manipulation in man. *Neuroscience Letters*, *323*(2), 113–116. https://doi.org/10.1016/S0304-3940(02)00119-2

Pfurtscheller, G. (1989). Functional topography during sensorimotor activation studied with event-related desynchronization mapping. *Journal of Clinical Neurophysiology*. https://doi.org/10.1097/00004691-198901000-00003

Pfurtscheller, G, & Lopes, F. H. (1999). Event-related EEG / MEG synchronization and desynchronization : basic principles. *Clinical Neurophysiology*, *110*, 1842–1857. https://doi.org/10.1016/S1388-2457(99)00141-8

Pfurtscheller, Gert, Zalaudek, K., & Neuper, C. (1998). Event-related beta synchronization after wrist, finger and thumb movement. *Electroencephalography and Clinical Neurophysiology - Electromyography and Motor Control*, *109*(2), 154–160. https://doi.org/10.1016/S0924-980X(97)00070-2

Pogosyan, A., Gaynor, L. D., Eusebio, A., & Brown, P. (2009). Boosting Cortical Activity at Beta-Band Frequencies Slows Movement in Humans. *Current Biology*, *19*(19), 1637–1641. https://doi.org/10.1016/j.cub.2009.07.074

Poldrack, R. A., Mumford, J. A., & Nichols, T. E. (2011). *Handbook of functional MRI data analysis*. Cambridge University Press.

Polyanska, L., Critchley, H. D., & Rae, C. L. (2017). Centrality of prefrontal and motor preparation cortices to Tourette Syndrome revealed by meta-analysis of task-based neuroimaging studies. *NeuroImage: Clinical*, *16*(July), 257–267. https://doi.org/10.1016/j.nicl.2017.08.004

Power, J. D., Barnes, K. A., Snyder, A. Z., Schlaggar, B. L., & Petersen, S. E. (2012). Spurious but systematic correlations in functional connectivity MRI networks arise from subject motion. *NeuroImage*, *59*(3), 2142–2154. https://doi.org/10.1016/j.neuroimage.2011.10.018

Rajagopal, S., Seri And, S., & Cavanna, A. E. (2013). Premonitory urges and sensorimotor processing in Tourette syndrome. *Behavioural Neurology*, *27*(1), 65–73. https://doi.org/10.3233/BEN-120308

Recio, G., Shmuilovich, O., & Sommer, W. (2014). Should I smile or should I frown? An ERP study on the voluntary control of emotion-related facial expressions. *Psychophysiology*, *51*(8), 789–799. https://doi.org/10.1111/psyp.12220

Reese, H. E., Scahill, L., Peterson, A. L., Crowe, K., Woods, D. W., Piacentini, J., … Wilhelm, S. (2014). The Premonitory Urge to Tic: Measurement, Characteristics, and Correlates in Older Adolescents and Adults. *Behavior Therapy*, *45*(2), 177–186. https://doi.org/https://doi.org/10.1016/j.beth.2013.09.002

Roberts, C. A., Giesbrecht, T., Stancak, A., Fallon, N., Thomas, A., & Kirkham, T. C. (2019). Where Is Itch Represented in the Brain, and How Does it Differ from Pain? An Activation Likelihood Estimation Meta-Analysis of Experimentally-Induced Itch. *Journal of Investigative Dermatology*. https://doi.org/10.1016/j.jid.2019.04.007

Roessner, V., Wittfoth, M., Schmidt-Samoa, C., Rothenberger, A., Dechent, P., & Baudewig, J. (2012). Altered motor network recruitment during finger tapping in boys with tourette syndrome. *Human Brain Mapping*, *33*(3), 666–675. https://doi.org/10.1002/hbm.21240

Salenius, S., Schnitzler, A., Salmelin, R., Jousmäki, V., & Hari, R. (1997). Modulation of Human Cortical Rolandic Rhythms during Natural Sensorimotor Tasks. *NeuroImage*, *5*(3), 221–228. https://doi.org/10.1006/nimg.1997.0261

SCAHILL, L., RIDDLE, M. A., McSWIGGIN-HARDIN, M., ORT, S. I., KING, R. A., GOODMAN, W. K., … LECKMAN, J. F. (1997). Children's Yale-Brown Obsessive Compulsive Scale: Reliability and Validity. *Journal of the American Academy of Child & Adolescent Psychiatry*, *36*(6), 844–852. https://doi.org/https://doi.org/10.1097/00004583-199706000-00023

Scharf, J. M., Miller, L. L., Gauvin, C. A., Alabiso, J., Mathews, C. A., & Ben-Shlomo, Y. (2015). Population prevalence of Tourette syndrome: A systematic review and meta-analysis. *Movement Disorders*, *30*(2), 221–228. https://doi.org/10.1002/mds.26089

Schultz, W. (1997). Dopamine neurons and their role in reward mechanisms. *Current Opinion in Neurobiology*, *7*(2), 191–197. https://doi.org/10.1016/S0959-4388(97)80007-4

Shute, J. B., Okun, M. S., Opri, E., Molina, R., Rossi, P. J., Martinez-Ramirez, D., … Gunduz, A. (2016). Thalamocortical network activity enables chronic tic detection in humans with Tourette syndrome. *NeuroImage: Clinical*, *12*, 165–172. https://doi.org/10.1016/j.nicl.2016.06.015

Silvestri, P. R., Chiarotti, F., Baglioni, V., Neri, V., Cardona, F., & Cavanna, A. E. (2016). Health-related quality of life in patients with Gilles de la Tourette syndrome at the transition between adolescence and adulthood. *Neurological Sciences*, *37*(11), 1857–1860. https://doi.org/10.1007/s10072-016-2682-y

Staba, R. J., Wilson, C. L., Bragin, A., & Fried, I. (2002). Quantitative Analysis of High-Frequency Oscillations (80 – 500 Hz) Recorded in Human Epileptic Hippocampus and Entorhinal Cortex,

(August 2015). https://doi.org/10.1152/jn.00322.2002

Steinberg, T., Shmuel Baruch, S., Harush, A., Dar, R., Woods, D., Piacentini, J., & Apter, A. (2010). Tic disorders and the premonitory urge. *Journal of Neural Transmission*, *117*(2), 277–284. https://doi.org/10.1007/s00702-009-0353-3

Stern, E, Silbersweig, D. A., Chee, K. Y., Holmes, A., Robertson, M. M., Trimble, M., … Dolan, R. J. (2000). A functional neuroanatomy of tics in Tourette syndrome. *Arch Gen Psychiatry*, *57*(8), 741–748. https://doi.org/10.1001/archpsyc.57.8.741

Stern, Emily, Silbersweig, D. A., Chee, K.-Y., Holmes, A., Robertson, M. M., Trimble, M., … Dolan, R. J. (2000). A Functional Neuroanatomy of Tics in Tourette Syndrome. *Archives of General Psychiatry*, *57*(8), 741. https://doi.org/10.1001/archpsyc.57.8.741

Storch, E. a, Murphy, T. K., Geffken, G. R., Sajid, M., Allen, P., Roberti, J. W., & Goodman, W. K. (2005). Reliability and validity of the Yale Global Tic Severity Scale. *Psychological Assessment*, *17*(4), 486–491. https://doi.org/10.1037/1040-3590.17.4.486

Thies, M., Zrenner, C., Ziemann, U., & Bergmann, T. O. (2018). Sensorimotor mu-alpha power is positively related to corticospinal excitability. *Brain Stimulation*, *11*(5), 1119–1122. https://doi.org/10.1016/j.brs.2018.06.006

Thomalla, G., Jonas, M., Bäumer, T., Siebner, H. R., Biermann-Ruben, K., Ganos, C., … Münchau, A. (2014). Costs of control: Decreased motor cortex engagement during a Go/NoGo task in Tourette's syndrome. *Brain*, *137*(1), 122–136. https://doi.org/10.1093/brain/awt288

Thut, G., Veniero, D., Romei, V., Miniussi, C., Schyns, P., & Gross, J. (2011). Rhythmic TMS causes local entrainment of natural oscillatory signatures. *Current Biology*, *21*(14), 1176–1185. https://doi.org/10.1016/j.cub.2011.05.049

Tinaz, S., Malone, P., Hallett, M., & Horovitz, S. G. (2015). Role of the right dorsal anterior insula in the urge to tic in Tourette syndrome. *Movement Disorders : Official Journal of the Movement Disorder Society*, *30*(9), 1190–1197. https://doi.org/10.1002/mds.26230

Van Der Salm, S. M. A., Tijssen, M. A. J., Koelman, J. H. T. M., & Van Rootselaar, A. F. (2012). The bereitschaftspotential in jerky movement disorders. *Journal of Neurology, Neurosurgery and Psychiatry*, *83*(12), 1162–1167. https://doi.org/10.1136/jnnp-2012-303081

van der Salm, S. M. A., van der Meer, J. N., Cath, D. C., Groot, P. F. C., van der Werf, Y. D., Brouwers, E., … Tijssen, M. A. J. (2018). Distinctive tics suppression network in Gilles de la Tourette syndrome distinguished from suppression of natural urges using multimodal imaging. *NeuroImage: Clinical*, *20*(August), 783–792. https://doi.org/10.1016/j.nicl.2018.09.014

Veniero, D., Vossen, A., Gross, J., & Thut, G. (2015). Lasting EEG/MEG Aftereffects of Rhythmic Transcranial Brain Stimulation: Level of Control Over Oscillatory Network Activity. *Frontiers in Cellular Neuroscience*, *9*(December). https://doi.org/10.3389/fncel.2015.00477

Wang, J., Wells, C., & Wu, L. (2011). NIH Public Access, *381*(1), 143–154. https://doi.org/10.1016/j.virol.2008.08.028.Macropinocytosis

Wang, Q., Chen, C., Cai, Y., Li, S., Zhao, X., Zheng, L., … Xue, G. (2016). Dissociated neural substrates underlying impulsive choice and impulsive action. *NeuroImage*, *134*, 540–549. https://doi.org/10.1016/j.neuroimage.2016.04.010

Wendel, K., Väisänen, O., Malmivuo, J., Gencer, N. G., Vanrumste, B., Durka, P., … Grave De Peralta Menendez, R. (2009). EEG/MEG source imaging: Methods, challenges, and open issues. *Computational Intelligence and Neuroscience*, *2009*. https://doi.org/10.1155/2009/656092

Woods, D. W., Piacentini, J., Himle, M. B., & Chang, S. (2005). Premonitory Urge for Tics Scale (PUTS): Initial psychometric results and examination of the premonitory urge phenomenon in youths with tic disorders. *Journal of Developmental and Behavioral Pediatrics*, *26*(6), 397–403. https://doi.org/10.1097/00004703-200512000-00001

Worbe, Y., Malherbe, C., Hartmann, A., Pélégrini-Issac, M., Messé, A., Vidailhet, M., … Benali, H. (2012). Functional immaturity of cortico-basal ganglia networks in Gilles de la Tourette syndrome. *Brain*, *135*(6), 1937–1946. https://doi.org/10.1093/brain/aws056

Worbe, Y., Marrakchi-Kacem, L., Lecomte, S., Valabregue, R., Poupon, F., Guevara, P., … Poupon, C. (2015). Altered structural connectivity of cortico-striato-pallido-thalamic networks in Gilles de la Tourette syndrome. *Brain*, *138*(2), 472–482. https://doi.org/10.1093/brain/awu311

Wright, A., Rickards, H., & Cavanna, A. E. (2012). Impulse-control disorders in gilles de la tourette syndrome. *The Journal of Neuropsychiatry and Clinical Neurosciences*, *24*(1), 16–27. https://doi.org/10.1176/appi.neuropsych.10010013

Xu, M., Kobets, A., Du, J.-C., Lennington, J., Li, L., Banasr, M., … Pittenger, C. (2015). Targeted ablation of cholinergic interneurons in the dorsolateral striatum produces behavioral manifestations of Tourette syndrome. *Proceedings of the National Academy of Sciences of the United States of America*, *112*(3), 893–898. https://doi.org/10.1073/pnas.1419533112

Yin, H. H., & Knowlton, B. J. B. B. J. (2006). The role of the basal ganglia in habit formation. *Nature Reviews. Neuroscience*, *7*(6), 464–476. https://doi.org/10.1038/nrn1919

Yin, S., Liu, Y., & Ding, M. (2016). Amplitude of Sensorimotor Mu Rhythm Is Correlated with BOLD from Multiple Brain Regions: A Simultaneous EEG-fMRI Study. *Frontiers in Human Neuroscience*, *10*(July), 1–12. https://doi.org/10.3389/fnhum.2016.00364

Zapparoli, L, Porta, M., Gandola, M., Invernizzi, P., Colajanni, V., Servello, D., … Paulesu, E. (2016). A functional magnetic resonance imaging investigation of motor control in Gilles de la Tourette syndrome during imagined and executed movements. *European Journal of Neuroscience*, *43*(4), 494–508. https://doi.org/10.1111/ejn.13130

Zapparoli, Laura, Macerollo, A., Joyce, E. M., Martino, D., & Kilner, J. M. (2019). Voluntary tic suppression and the normalization of motor cortical beta power in Gilles de la Tourette syndrome: an EEG study. *European Journal of Neuroscience*, (April), 1–14. https://doi.org/10.1111/ejn.14548

Appendix 1

Participants

HS: 12 participants (6 females) between 19 and 28 (mean age of 21.8) were recruited from the University of Nottingham. One participant was left-handed.

TS: 14 participants (5 females) between 12 and 33 (mean age of 20.3) were recruited for the study. Three participants were left-handed.

Event related perturbation (ERP) in HS

The ERP showed the readiness potential (RP) and motor potential (MP) that characterises the voluntary movement (see Figure 8.1). The RP starts at 535ms before the MP. The MP starts with the button press and ends 355ms after that. This is followed by a post-movement rebound.

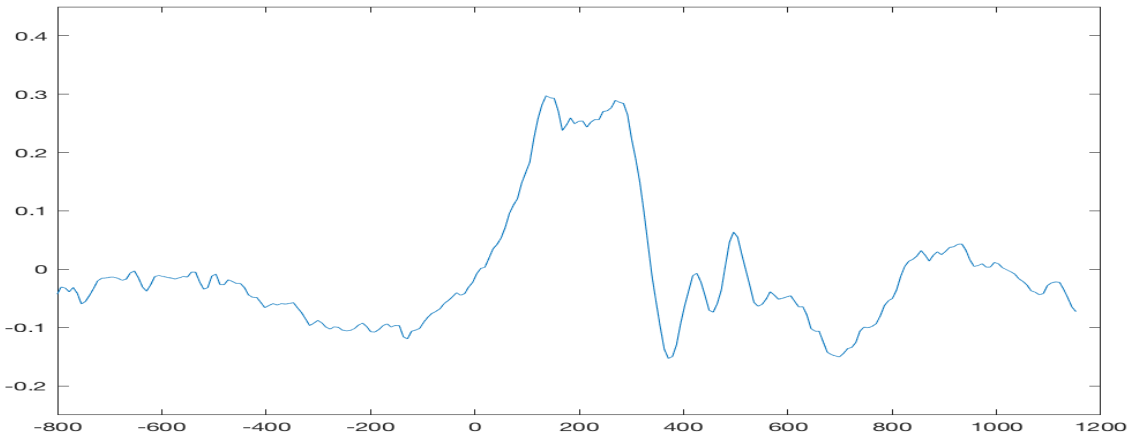


Figure 8.65. ERP shown in the contralateral M1. Time 0 indicates the start of the button press.

Event related perturbation (ERP) in TS

The ERP showed the readiness potential (RP) and motor potential (MP) that characterises the voluntary movement (see Figure 8.2). The RP starts at 535ms before the MP. The MP starts with the button press and ends 347ms after that. This is followed by a post-movement rebound.

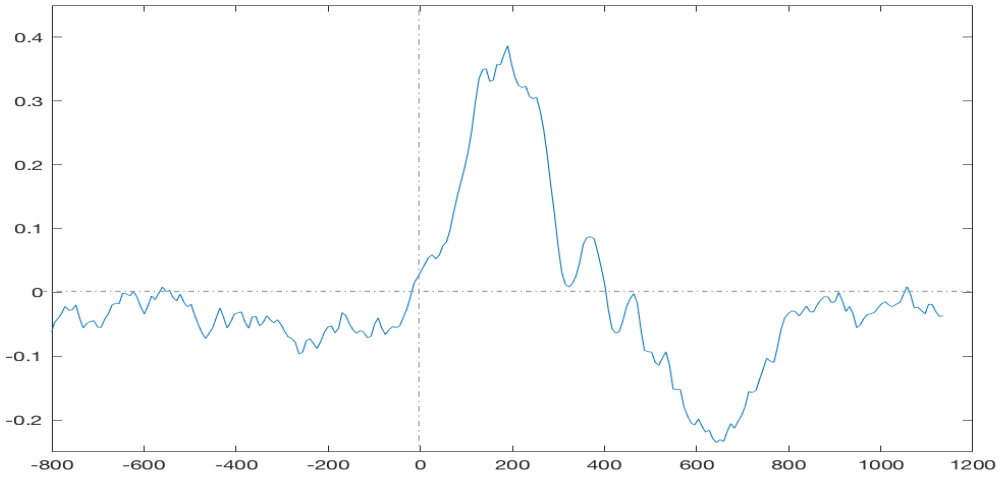


Figure 8.66. ERP shown in the contralateral M1. Time 0 indicates the button press.

Mu rhythm in HS

The mu rhythm in the contralateral M1 showed a significant reduction in power (ERD) prior to the button press and during the execution of the finger movement (see Figure 8.3). The significant ERD at 12Hz starts 20ms before the movement and ends 370ms after the button press.

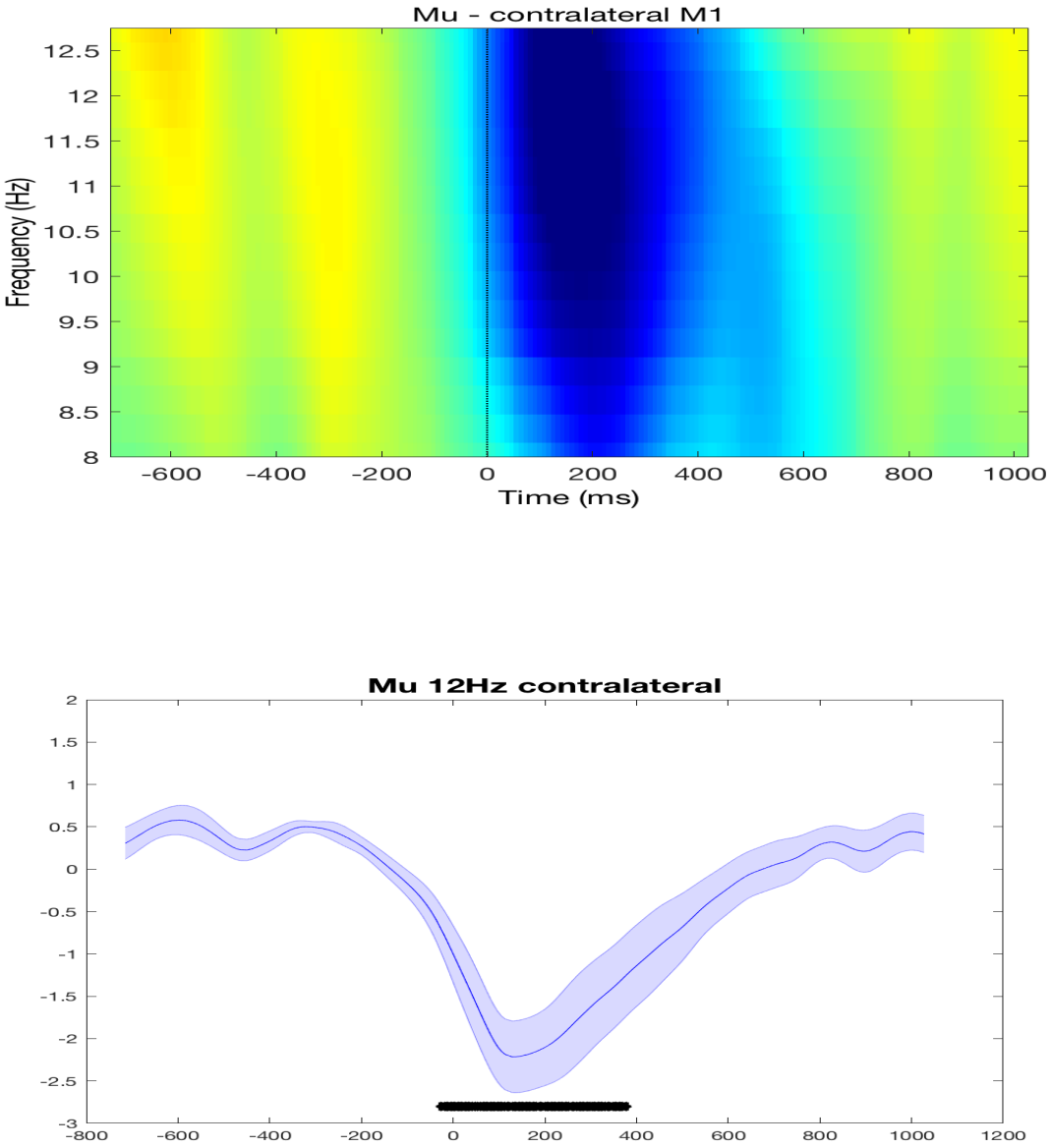


Figure 8.67. A: Time-frequency analyses from the contralateral M1 in the mu band; B: Values of power at 12Hz recorded from the contralateral M1 in time. Time 0 indicates the button press. The black horizontal line indicates the time points were the values of power are significant.

Mu rhythm in TS

The mu rhythm in the contralateral M1 showed no significant reduction in power (ERD) prior to the button press and during the execution of the finger movement (see Figure 8.4).

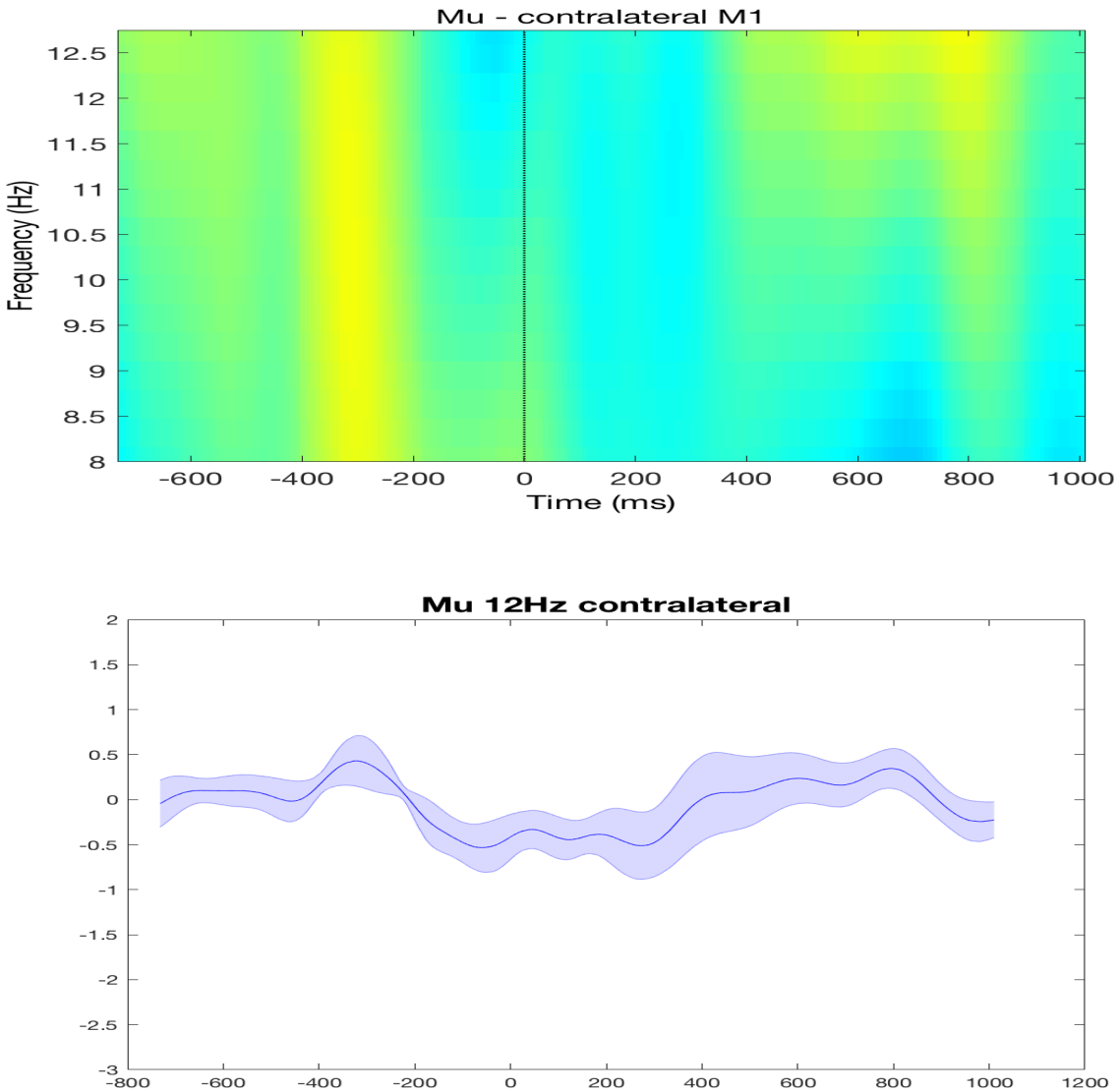


Figure 8.68. A: Time-frequency analyses from the contralateral M1 in the mu band; B: Values of power at 12Hz recorded from the contralateral M1 in time. Time 0 indicates the start of the movement.

Beta rhythm in HS

The beta rhythm in the contralateral M1 showed a significant reduction in power (ERD) before the button press and during the execution of the finger movement (see Figure 8.5). The significant ERD at 19Hz starts 191ms before the movement and ends 238ms after the button press.

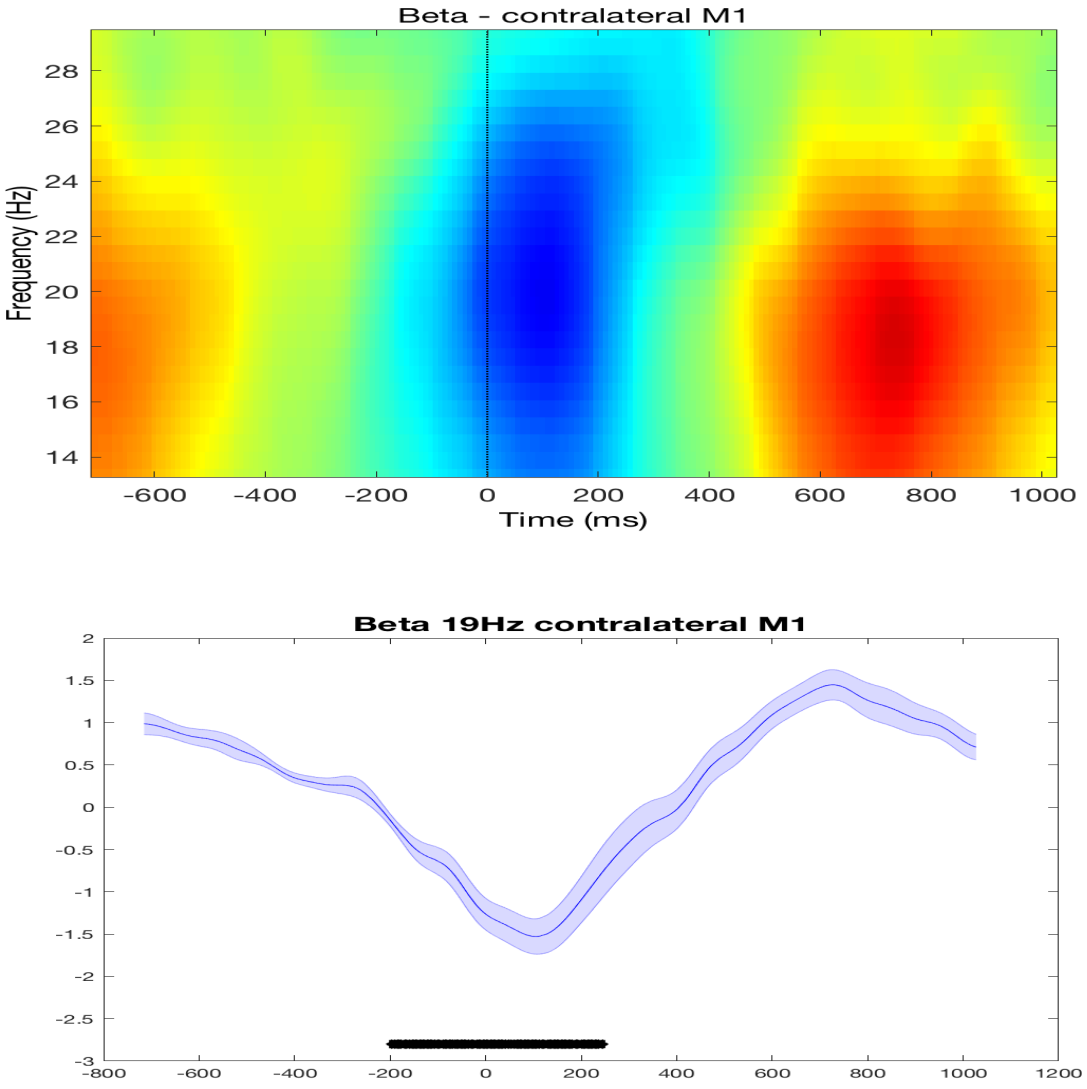


Figure 8.69. A: Time-frequency analyses from the contralateral M1 in the beta band; B: Values of power at 19Hz recorded from the contralateral M1 in time. Time 0 indicates the start of the movement. The black horizontal line indicates the time points were the values of power are significant.

Beta rhythm in TS

The beta rhythm in the contralateral M1 showed a significant reduction in power (ERD) before the button press and during the execution of the finger movement (see Figure 8.6). The significant ERD at 19Hz starts 113ms before the movement and ends 66ms after the start of the movement.

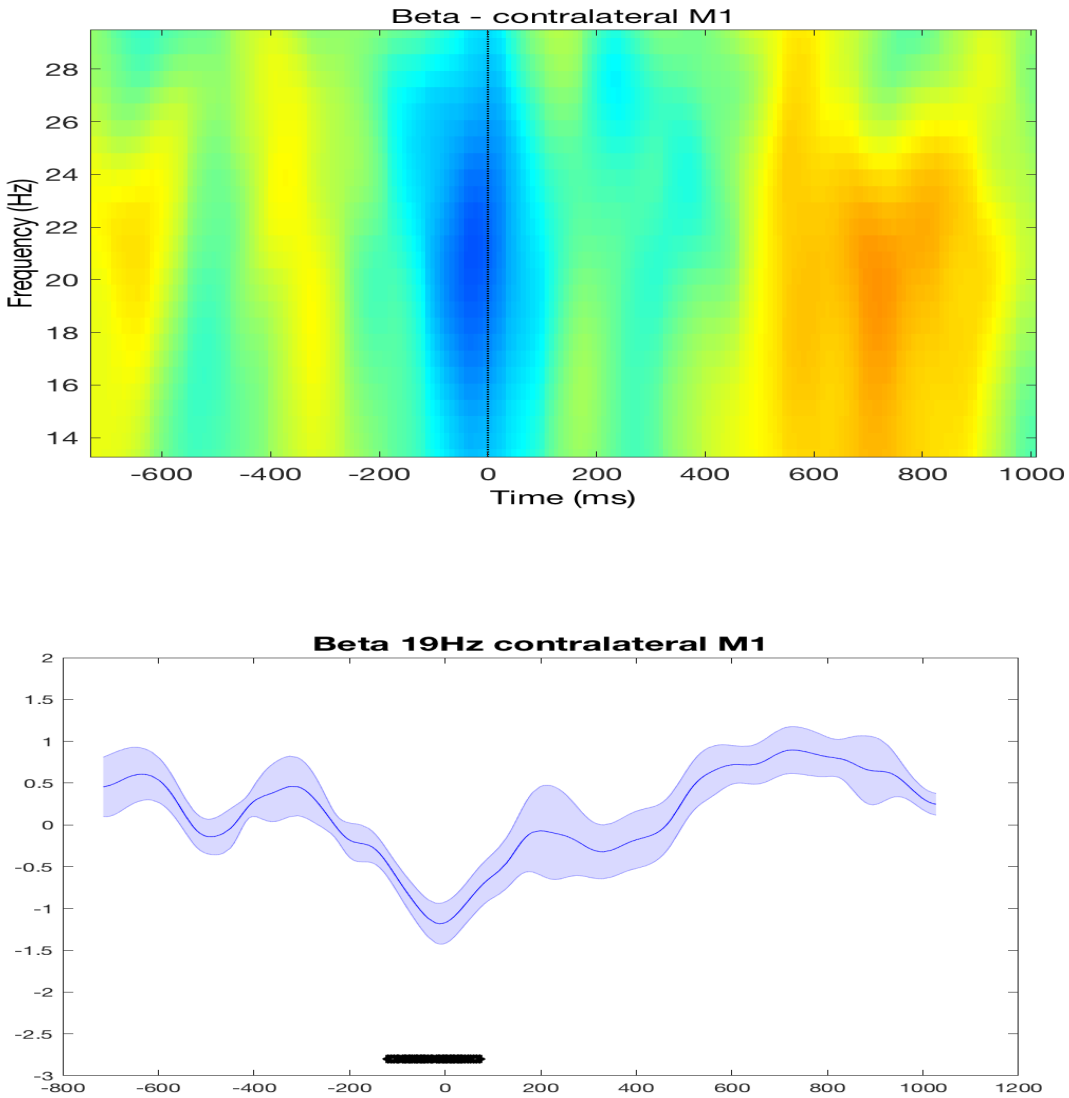


Figure 8.70. A: Time-frequency analyses from the contralateral M1 in the beta band; B: Values of power at 19Hz recorded from the contralateral M1 in time. Time 0 indicates the start of the movement. The black horizontal line indicates the time points were the values of power are significant.

Appendix 2

Figure 9.1 shows the time-frequency plots in alpha, beta, and gamma, when filtering from 8-63Hz for the average electrodes on the SMC. Results show a strong ERS starting shortly before tic onset that lasts for the rest of the epoch and includes all alpha, beta, and gamma bands.

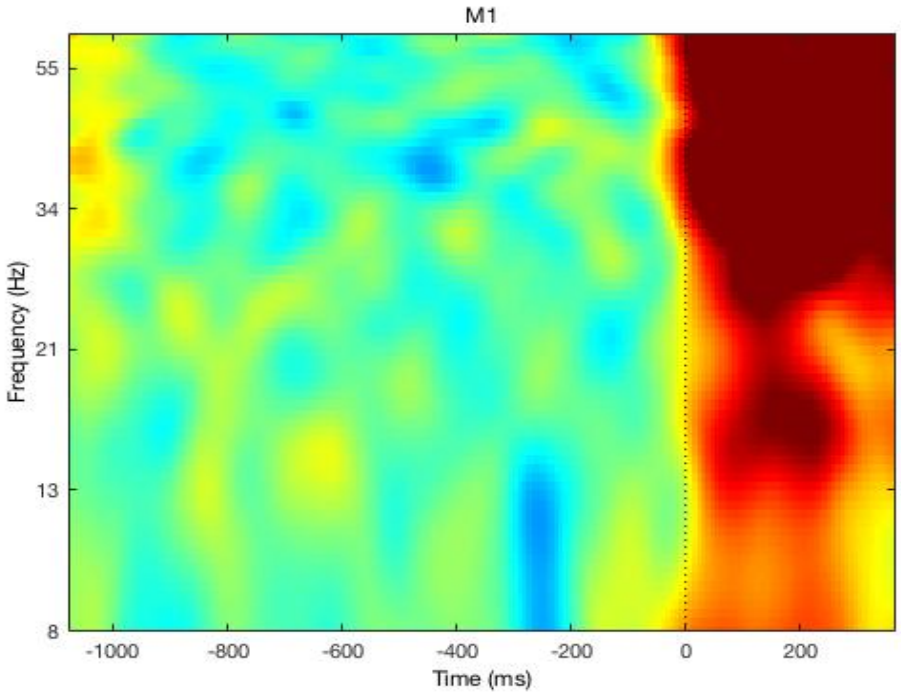


Figure 9.71. Time-frequency plots in alpha, beta and gamma bands (8-63Hz) for the average electrodes on the SMC. 0 indicates the onset of the tic.